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THE GENUS *TILIA* IN OHIO*

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Examination of about 250 Ohio specimens of *Tilia* shows that the Ohio population consists of three basic species (*americana* L., *heterophylla* Vent., *floridana* [V. Engler] Small) and a species-complex here referred to as *T. neglecta* Spach.

T. americana varies considerably; the limits of the taxon are obscure, not only in Ohio, but through much of its range. However, in the northwestern part of its range, in Minnesota, the species seems more stable than elsewhere; little variation was observed upon examination of a considerable number of specimens (in University of Minnesota herbarium). Some Ohio specimens resemble those of this northwestern population; others suggest introgression from some other species, probably *heterophylla*. The glacial and postglacial history of *Tilia* furnishes a clue to the explanation.

At the time of the last (Wisconsin) ice maximum, *Tilia* survived only beyond the ice margin—in the Driftless Area of Wisconsin, a great re-entrant angle between glacial lobes; on the northern Appalachian Upland; and elsewhere in front of the ice border, which lay across Indiana and Ohio. In its postglacial migration, as indicated by pollen analysis of bogs (Sears, 1942), *Tilia* appeared early in what is now the northwestern part of its range, earlier than (with few exceptions) in the central part of its range (Indiana, Ohio). These locations of early entry into Ohio and Indiana are north of bogs whose records show later entry, and are near the periphery of the Prairie Peninsula. This suggests spread from a refugium in Wisconsin (and possibly farther to the southwest), spread not only westward, but also southeastward. This *Tilia* population had not come in contact with other taxa, hence is the least variable and the true basic species (even if it does not coincide with a taxonomic type).

Comparison of Ohio material with northwestern material shows that some specimens resemble the more northwestern ones. These are shown on the map (fig. 2 A) by dots with an upright appendage, and are here considered to represent the basic species, uncontaminated by other species (fig. 1 A). Many other specimens are sufficiently similar to be placed in the same taxonomic "pigeon-hole"; these are shown by dots on the map. Still others are generally referred to *T. americana*, but depart in one or more characters from the basic taxon; these are shown by circles on the map. *T. americana* (inclusive) is, in Ohio, almost confined to the area of Wisconsin glaciation.

T. heterophylla (fig. 1 B) is found in unglaciated Ohio and the dissected border of Illinoian glaciation (fig. 2 B). It is a characteristic species of the Mixed Mesophytic climax association, occurring abundantly in that association in the Cumberland Mountains and Cumberland Plateau and in coves of the Great Smoky

*Study of Ohio specimens made as part of work on "Woody Plants of Ohio" and supported by a grant from the National Science Foundation.

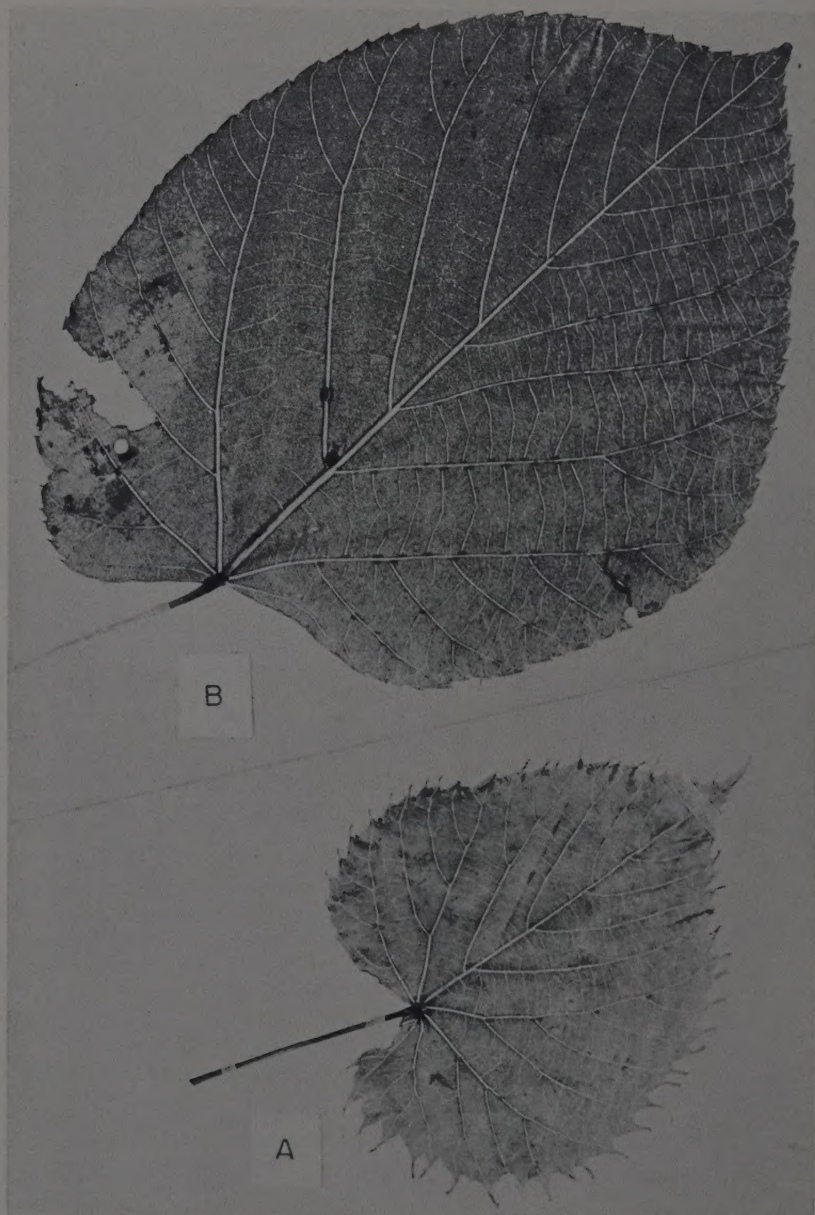


FIGURE 1. A, Leaf of *T. americana*. B, Leaf of *T. heterophylla*, natural size. (From photo-stats of pressed leaves.)

Mountains. An abundant species of the old (ancestral) area of deciduous forest, and probably by Pleistocene time almost confined to the Appalachian Highland, its range later must have been curtailed on the north by Pleistocene ice. *T. floridana*, represented by a few specimens from unglaciated eastern Ohio, may have

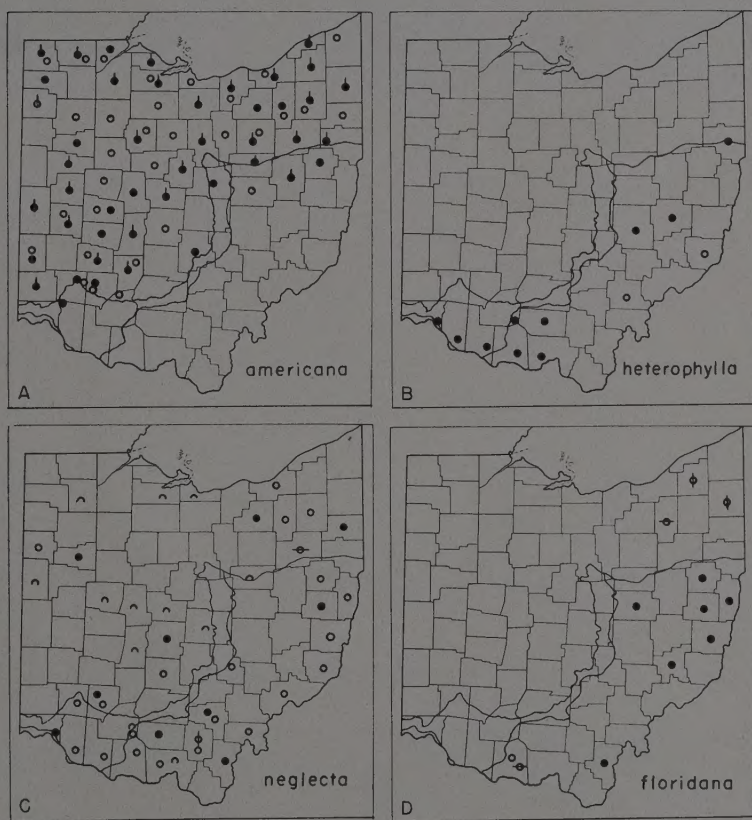


FIGURE 2. A, B, C, D. Distribution of four species of *Tilia* in Ohio.

had a similar history. Along a tension zone south of the Wisconsin ice, unlike races of *Tilia* met and mixed, resulting in introgression from *T. heterophylla* (and probably *T. floridana*) into the more northern *T. americana*.

In order to analyze variation as displayed by the Ohio population of *Tilia*, pictorialized scatter-diagrams of the type devised by Anderson (1949, 1952, 1953) have been constructed (fig. 3). For plotting *americana*, only specimens representing the "basic species" were used. The glyphs of this species occupy a definite and circumscribed area on the diagram, and are far-removed from those of *heterophylla*. Glyphs of a third "species" or "species-complex," *T. neglecta*, occupy an

intermediate position, overlapping in position those of *americana* and *heterophylla*.

Basic *T. americana* may be recognized by three leaf characters: leaves glabrous beneath, with axillary tufts at primary veins, rarely at secondary veins; large jagged teeth usually 3-5 mm high; few teeth per unit length, 3-5 in 2 cm of leaf margin. *T. heterophylla* differs markedly in these three characters: leaves densely pubescent beneath with matted branched hairs, axillary tufts present but usually not conspicuous; teeth low, less than 2 mm high; more teeth per unit length, usually 6-8 or more in 2 cm of leaf margin. *T. neglecta*, as here used, lacks such definite characters; leaves vary from almost glabrous to thinly pubescent with

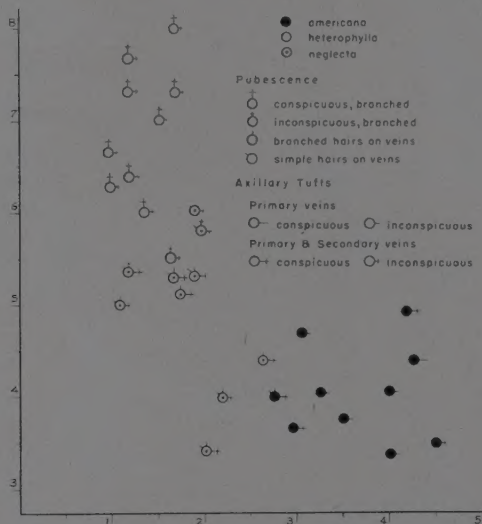


FIGURE 3. Pictorialized scatter-diagram of Ohio population of *Tilia americana*, *heterophylla*, and *neglecta*, based on leaf characters. Horizontal axis: height in mm of upper (apical) edge of teeth. Vertical axis: number of teeth per 2 cm of leaf margin. Glyph appendages indicate pubescence and axillary tufts; differences in length relate to intensity of character depicted.

simple or branched hairs or both; axillary tufts usually prominent; teeth vary in size, shape, and in spacing. In certain characters, it resembles *americana*, in others, *heterophylla*. Features of the glyphs and their positions indicate that *T. neglecta* has arisen postglacially as a result of contact between *americana* and *heterophylla*.

Distribution of *T. neglecta* (dots on map), of specimens doubtfully referred to *neglecta* (circles on map), and of those which might equally well be thought of as *americana* in part (half-circles on map), shows this complex (fig. 2 C) to be most abundantly represented near the zone of contact of *heterophylla* and *americana*, but present also farther north well within the range of *americana*. Hybridization and backcrossing with *americana* have carried introgression far northward. Occa-

sional specimens have characters suggesting *heterophylla* (horizontal line through circle) or *floridana* (vertical line through circle).

In some local areas just outside the glacial boundary, variation in the *Tilia* population suggests hybrid swarms. Such is the case with the *Tilia* population in Fort Hill State Memorial, Highland County, where some specimens are definitely referable to *T. heterophylla*, some doubtfully to *T. americana*, and others to *T. neglecta*. The scatter-diagram (fig. 4) shows the glyphs scattered from within the area of *americana* glyphs of fig. 3 to within the area of *heterophylla* glyphs, with most in the intermediate position of *T. neglecta*.

T. floridana (fig. 2 D) seems to have played a lesser part in the variation of *Tilia* in Ohio. A few specimens suggest admixture with *heterophylla* (horizontal

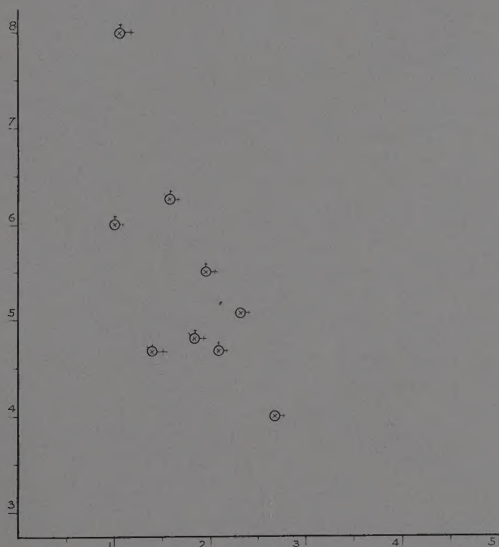


FIGURE 4. Pictorialized scatter-diagram of *Tilia* population of Fort Hill, Highland County. Axes and glyph appendages as in figure 3.

line through circle), a few (from the northern part of the Allegheny Plateau) with *americana* (vertical line through circle). It is so poorly represented, numerically, that no analysis is possible. The leaves are coarsely toothed as in *americana*; but the almost imperceptible pubescence of the lower leaf-surface, which imparts a velvety feel even when the leaves appear glabrous, will distinguish it from other species.

A key for the identification of these species of *Tilia* as seen in Ohio will be found in "The Woody Plants of Ohio" (Braun, 1960, Ohio State University Press).

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THE SUMMER FIELD MEETINGS OF THE OHIO ACADEMY OF SCIENCE (1892-1905)

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The Ohio Academy of Science was formally created at an organizational meeting held in Columbus on December 31, 1891. One of its first projects was to sponsor a series of annual field meetings. These ran continuously from 1892 until 1901. The last one was held, after a brief interval, in the summer of 1905. The field meetings were initiated even before the first of the regular annual business meetings was held. During the early years of the Academy, the summer field meeting was considered one of its most important activities. This paper will trace the history of those events.

The first Annual Field Meeting, of what was then called the Ohio State Academy of Science, was held at Akron on June 3 and 4, 1892. This meeting was sponsored by the Science Department and the Akron Scientific Club of the institution known at that time as Buchtel College (later this became the University of Akron). At this initial meeting, there was a brief business session even though the first annual meeting of the Academy was not called until November of that year. Friday afternoon was given to field study and collecting at Long Lake, on which the group cruised about on a steamer. This trip was arranged for those interested in "geology, botany, zoology, entomology, ornithology, conchology," In the evening Professor E. W. Claypole, the first president of the Academy, had been scheduled to give an address of welcome in Crouse Gymnasium, but the mayor of Akron and Dr. O. Cone, President of Buchtel College, were finally asked to do this. The words of Dr. Cone reflected a concern at that time held by many who were worried about the introduction of science teaching into the college curriculum. Dr. Cone said in part, "We have no fear of science at Buchtel College. We are not afraid that its study will endanger a single truth. There is not a truth it has touched that it has not illuminated. Science enlarges most views—the marvelous truth of evolution has only brought into belief the greatness of the first cause." Following the speeches, a discussion of the observations made during the afternoon's excursion was enjoyed. Students of Buchtel College placed on exhibition microscopical slides which they had prepared. Arrangements were made to continue the field work for the following day in the gorge at Cuyahoga Falls where the glens held much of interest to the various naturalists.

In the spring of 1893 the executive committee mailed a circular announcing the second summer field meeting to be held at Logan in Hocking County on June 2 and 3. During the first morning a visit was made to the Logan Pressed Brick and Tile Works to observe the processes of manufacture. During the afternoon a field trip on the Hocking River gave the specialists an opportunity to collect specimens. That evening a meeting was held in the Presbyterian Church of Logan at which time President Edward Orton presided and presented a brief summary of the geology of that region. Professor W. A. Kellerman then called attention to the special botanical interests in that area. Dr. Clark placed on exhibition a fossil jaw which he had collected; Dr. Claypole commented upon the specimen and designated it as an unnamed species of *Dinichthys*; and Dr. Kellicott reported briefly on the activities of the entomologists who visited Cedar Swamp. It was at this meeting that plans were initiated for the development of a Natural History Survey.

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On June 3, members of the Academy went to Straitsville to visit a coal mine. On the way some of the entomologists and botanists left the party to engage in field collecting. The geologists and others who were conducted through the mine collected coal measure fossils.

The third field meeting was held June 1 and 2, 1894, at Denison University in Granville. One party spent a full day collecting at Licking Reservoir while another spent the day in field study around Granville. The new Science Hall of the university attracted much attention, and under the guidance of Professor W. G. Tight a visit was made to observe serious soil and river bank erosion. A series of photographs depicted the annual progression of erosion. The geologists collected fossils at the Waverly quarries. In the evening the Scientific Association of Denison University entertained the group with informal discussions and the projection of microscope slides following brief remarks by President Purinton and Professor F. M. Webster of Denison University and several prominent members of the Academy. On Saturday morning a group of 60 people visited the Effigy Mound on Alligator Hill and other ancient earthworks and archeological remains in the vicinity of Newark. A visit was also made to a postglacial gorge of the Licking River some twelve miles east of Newark, where these gorges were studied particularly by the geologists in the party under the direction of Professor Tight.

For the 1895 field meeting the Academy met at Sandusky in early July, meeting jointly with the Ohio Teachers' Association. Arrangements were made by Professor E. L. Moseley who also conducted the field excursions. A trip was first made to Cedar Point where a special study was made of the sand dunes, the cacti and other sand plants, and reptiles found in that area. The archeologists were taken to an old flint quarry and Indian arrow factory a few miles south of Sandusky. A visit was also made to the "Blue Hole" at Castalia and a neighboring old-lake ridge. Professor D. S. Kellicott, President of the Academy, explained the nature of the Academy to the Ohio Teachers Association during the evening meeting. W. H. Todd presented a paper on "A Cyclone in Erie County," and Dr. G. F. Wright gave the principal address of the evening, entitled "Evidences of the Glacial Age in Ohio."

The following day was devoted to a field excursion which took members of both organizations to Marblehead and three of the offshore islands (Sugarloaf, Green, and Kelley's Islands). At Marblehead and on Kelley's Island the glacial grooves were examined under the direction of G. F. Wright.

The field meeting of 1896 was a joint conclave of the Ohio and Indiana Academies of Science, held at Oxford on June 4 and 5. Miami University, Western College for Women, and Oxford Female Seminary served as hosts. The geologists made a trip to Darrrtown to observe glacial till; paleontologists collected trilobites at High Bank; biologists collected in local ponds and creeks; and anthropologists examined an Indian mound. On Thursday evening a banquet was held at Western College. An address by Professor Stanley Coulter of DePauw University was given in the chapel of Miami University. Friday evening dinner was served at the Oxford Female Seminary followed by an illustrated address given in Miami Chapel by R. E. Call on the subject of "Mammouth Cave." Professor A. L. Treadwell of Miami University was in charge of the local arrangements, and he provided visitors with collecting equipment and photographic supplies.

The sixth annual summer meeting was held at Brinkhaven and Gambier under the auspices of Kenyon College. Glacial moraines and tills were explored by the geologists, and the Mohican River was examined as far north as Alum Rocks by the various naturalists present. Following the field excursion at Brinkhaven, the group assembled at Kenyon College where an evening meeting was held. Following an address of welcome by President Pierce of Kenyon College and a response given by Professor E. W. Claypole, Miss Mary Hart of Western College spoke on "The Education of Women" and Mrs. W. A. Kellerman reported on the Women's

National Science Association. Mrs. Claypole described the scientific careers of her daughters who were professors at Wellesley College and Professor W. G. Tight then gave an illustrated lecture on past and present drainage in the Gambier area. On Saturday morning a field trip was made under the Direction of W. A. Kellermar to an area along Owl Creek known as the "Caves."

In 1898 the field meeting was held at Dayton on June 3 and 4. A visit was made to the glacial region south of Dayton on Friday afternoon, and that evening an illustrated lecture on glaciers was delivered by Dr. A. F. Foerste. On Saturday morning the naturalists went to Yellow Springs and to Clifton Gorge, which is cut through upper Silurian limestone, for observations and collecting activities. The Dayton science teachers and the science professors and students of Antioch College assisted with the arrangements.

The summer meeting for 1899 was held in Columbus as a part of the meeting of the American Association for the Advancement of Science. Because of the larger meeting in session, the Academy group did little more than hold a business meeting and hear an address by Rev. Herzer on the fossil genus *Sigillaria*.

The ninth summer meeting was held at Put-in-Bay on South Bass Island in Lake Erie from June 26-28. The Ohio Teachers Association also held its meeting at the same time and place. A visit was made to Rattlesnake Island and Middle Bass Island. The lake was too rough to continue to either Marblehead or Kelley's Island as had been planned. The group did, however, visit the U. S. Fish Hatchery, Crystal Cave, and Daussa Cave. At this meeting it was announced that arrangements had been made through the Ohio Legislature to engage the U. S. Geological Survey to undertake a topographic survey of the State of Ohio. This project, under the guidance of A. A. Wright of Oberlin, was considered the most important achievement of the Academy to that date.

The last regular field meeting since the founding of the Academy took place on May 31 and June 1, 1901, at Wooster. The University of Wooster (now the College of Wooster), the Ohio Agricultural Experiment Station, and the Wooster Field-Naturalist's Club served as hosts. The bog and adjoining woods around Brown's Lake, located west of Shreve, were visited by the naturalists. A visit was made to the Agricultural Experiment Station and a trip was made up the Killbuck Valley to visit coal mines and Fox Lake with its tamarack bog. It was announced that special paper number 4 on "The Fishes of Ohio" was nearly ready for publication.

At the annual meeting held in Columbus that year, the secretary reported, "In view of the fact that the attendance at the field meetings for the last six years has been small except when held in connection with the meeting of some other association, I recommend that we abandon the policy of holding a field meeting every year, and without fixing a longer period, leave the matter to be decided each year by the executive committee with power to act in making future plans for Academy meetings."

The final independent field meeting of the Academy was held four years later at "The Breakers" and the Lake Laboratory of Ohio State University situated at Cedar Point near Sandusky, July 5 to 8, 1905. A meeting of the American Microscopical Society was held at the same place and at that time, making it possible for Academy members to attend both meetings. It was the 28th annual meeting for the American Microscopical Society, and it was upon its invitation that this joint meeting was planned. As in the past, much of the time was spent in field collecting. Visits were made to Johnson Island, Kelley's Island, and Put-in-Bay. A part of the joint program was a symposium on "Animals as Parasites and Carriers of Disease" and a symposium on "Freshwater Biology." Participants included E. A. Birge, L. B. Walton, C. H. Eigenmann, Herbert Osborn, R. H. Wolcott, F. L. Landacre, and Henry B. Ward.

No other separate field meeting has been held by the Ohio Academy. Some

of the state academies, notably the Indiana Academy of Science, have continued field meetings over many years time. The Ohio Academy has, however, often scheduled field trips immediately following the regular annual meeting. Field excursions were held in 1910, 1918, 1919, 1925, 1928, 1931, 1934, 1936, 1938, and regularly since 1952. These have been sponsored for the most part by the sections of zoology, plant sciences, geology, and conservation. Occasionally the chemistry, physics, and geography sections have sponsored visits to industrial plants and similar organizations located near the place of the annual meeting. Many of the field trips in recent years have been made to the same localities visited in the earlier days. To some members of the Academy the opportunity to visit natural areas throughout the State of Ohio and to make field collections have been high-lights at the meetings of the Academy.

Acknowledgment is made to Dr. Dwight M. DeLong, President, and Dr. George W. Burns, Secretary, of The Ohio Academy of Science for a critical reading of the manuscript.

PENTREMITIDEA FILOSA WHITEAVES FROM THE SILICA FORMATION IN NORTHWESTERN OHIO

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INTRODUCTION

Blastoids were described from the Silica formation in northwestern Ohio by Kier (1952). Two species, *Nucleocrinus* sp. cf. *N. elegans* Conrad and *Pentremitidea reimanni* Kier, were collected from unit 13 of the Silica formation, as designated by Ehlers, Stumm and Kesling (1951), from the south quarry of the Medusa Portland Cement Company west of Sylvania, Ohio. The specimens described here were collected from unit 6 (Ehlers, Stumm and Kesling, 1951) in the north quarry of the same company. The north quarry is located approximately three miles southwest of Sylvania, Ohio, on the north side of Brint Road just west of Centennial Road in the SE $\frac{1}{4}$ sec. 7, T. 9 S., R. 6 E.

SYSTEMATIC DESCRIPTION

ORDER EUBLASTOIDEA

Family Pentremitidae

Genus *Pentremitidea* Orbigny, 1849

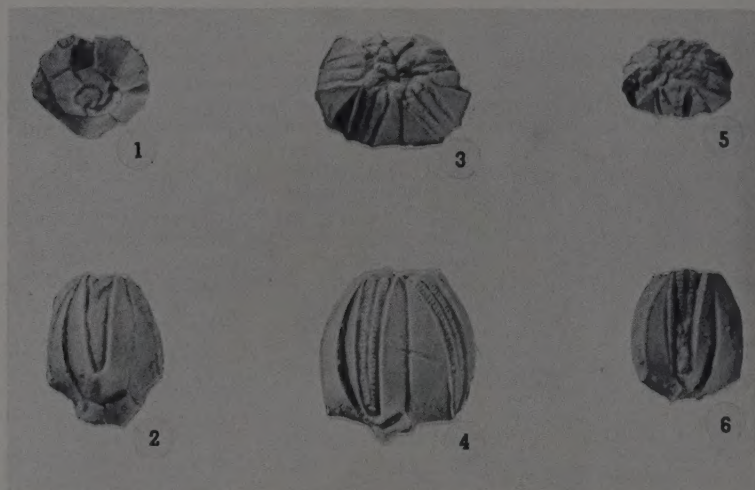
Pentremitidea filosa Whiteaves, 1889

Figures 1-6

Pentremitidea filosa Whiteaves, 1889, Contributions to Canadian Palaeontology, 1: 104-107, pl. 14, figs. 1, 1a, 1b.

Calyx of medium size with the undistorted specimen being 11.3 mm high and 8.1 mm wide. Greatest width below mid-height, usually at or near the base of the ambulacra. The edges of the radial plates adjacent to the ambulacral areas are raised forming a narrow ridge. Relative dimensions of radial plates and

ambulacral areas shown in table 1. Deltoids extremely small with the anal deltoid appreciably larger and rhomboid in shape. Three basal plates with one smaller than the others. Ambulacral areas narrow and widening uniformly towards the oral surface. Groove in the center of the ambulacrum is more pro-



EXPLANATION OF FIGURES IN PLATE

All figures on this plate are of *Pentremiteidea filosa* Whiteaves and are magnified $\times 2$.

1, 2. Basal and lateral views. BGSU No. 1051.

3, 4. Summit and lateral views. BGSU No. 1052.

5, 6. Summit and lateral views. BGSU No. 1053.

nounced towards the oral surface being faint near the bottom of the ambulacral area.

Ornamentation on the surface of the radial plates consists of very fine lirae which parallel the outer edge of the plate but which are subparallel near the cen-

TABLE 1
Comparison of radial and ambulacral dimensions.

Specimen	H.	W.	Length Radial	Width Radial	Length Amb.	L/W. Ratio Radial	Length Ratio Radial & Amb.
1	12.2*		11.7	6.8	11.0	.58	.940
2	11.3	8.1	9.8	5.0	8.9	.51	.908
3	9.8*		9.5	4.6	8.6	.48	.905

*Basal plates absent.

Dimensions in millimeters.

tral portion of a side of a plate. The lirae begin well below the oral apex of the radials with the longer ones near the outer edge and then becoming progressively shorter as the ambulacral area is approached. The lirae extend to the base of

the radials but were not seen on the basal plates. Brachioles and summit features not preserved.

Remarks.—The specimens, although partially crushed, agree closely with the original description by Whiteaves (1889), especially in the size relationships of the radial plates and ambulacral areas. However none of the basal plates in the specimens described here are quadrangular as he mentions.

Pentremilidea filosa differs from *P. goldringae* Reimann, *P. southworthi* Reimann, *P. cummingsi* Reimann and *P. claviformis* Reimann by having its greatest width closer to the base of the calyx and less attenuate basal plates. *P. perovale* Reimann has ambulacrals which expand more rapidly and although the basal plates are near the same size they comprise one-fifth of the total height in *P. perovale* and only one-seventh in the specimens described above. *P. americana* Barris has more widely expanding ambulacral areas and a longer more attenuate base (Barris, 1883). *P. reimanni* Kier has shorter ambulacral areas in relation to the length of the radial plates. *P. cooperi* Reimann has a rounded summit and *P. bellatula* Reimann has a smaller summit area than *P. filosa* (Reimann, 1935, 1940, 1945).

Apparently there is an increase in the ratio of the length of the radial plates and the length of the ambulacral area and also an increase in the length/width ratio of the radial plates in connection with an increase in total size (table 1). The fine surface markings also become more prominent as size increases.

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The Study of Rocks in Thin Section. W. W. Moorhouse. Harper & Brothers, New York. 1959. xvii+514 pp. \$8.00.

This admirable book is so written that it may serve as a text for the student, a manual for the practicing geologist, and a sort of first-reference source for both. A summary of the methods of optical mineralogy, descriptions of the plentiful rock-forming minerals (with 18 fine plates of microphotos), and tables for mineral identification are brought together with the petrographic and petrologic material. This mineralogic section occupies about 30% of the pages of the book. The rock groups, including all of the important ones, are arranged under four main sections—igneous (37%), sedimentary (13%), metamorphic (16%), and ores (3%). The altered rocks are grouped according to their general mode of alteration—dynamic, thermal, regional, metasomatic.

Both the arrangement and the content of each section concerning a clan of rocks are excellent, and the uniformly systematic treatment of topics is very helpful. This book is not designed to serve the specialist; its value is the juxtaposition of petrography and petrology, as well as its breadth and quality. The combination of petrography and petrology suggests Grout's text of that name, but only thin section petrography is included in the present book.

MALCOLM P. WEISS

MORTALITY OF FRESH WATER AND TROPICAL FISH FRY BY CYCLOPOID COPEPODS

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INTRODUCTION

Observations of attacks of cyclopoid copepods on larval fish have been described infrequently in the literature. Davis (1959) found dead fish fry from Lake Erie with *Cyclops bicuspidatus* and *Mesocyclops edax* attached. He found that a laboratory culture of *Mesocyclops edax* equivalent to a population density of 500 copepods per liter could damage the fins of young rockbass, *Ambloplites rupestris*. Fryer (1953) observed *Cyclops viridis* attacking a recently hatched fish. In the same account, he reported that an aquarist witnessed attacks by *Cyclops* sp. on newly hatched tropical fish fry and attributed the deaths of two dozen young to this cause. Oliva and Sládeček (1950) reported the death of a 2-cm long axolotl larva following attacks by *Cyclops strenuus* and *Cyclops vicinus*. Spandl (1926) states that cyclopoid copepods may attack and injure small fish, and that death possibly results from infection after the attack. Innes (1955) saw cyclopoids attached to young fishes.

The present paper describes experiments designed to confirm the observations of Davis and to test the effect of population density of cyclopoids and of various sizes of fish fry on predation rates. These experiments constitute the initial phase of a longer research program which will be an attempt to evaluate the role of cyclopoids as natural predators of young fish.

MATERIALS AND METHODS

Adult and copepodid stages of *Cyclops vernalis*, *C. bicuspidatus*, and *Mesocyclops leuckarti*, were collected near the Fisheries pier or in open water of Hatchery Bay, Put-in-Bay, Ohio. Copepods were usually collected on the day they were used to start experiments, but, in some instances when cyclopoids were impossible to obtain in quantity, cultured *C. vernalis* were used. Differences between the behavior of freshly caught animals and that of those maintained in laboratory cultures were not noted.

Fish eggs and fry were collected from nests in a small farm pond in Beaver County, Pennsylvania, and in Terwilliger's Pond, South Bass Island, Ohio. Bluegill fry, *Lepomis macrochirus*, were obtained from the nest area in the tanks at the Toledo Aquarium. Zebra eggs and fry of *Brachydanio rerio* (Hamilton-Buchanan), the fry of the gourami, *Trichogaster trichopterus*, and the eggs and fry of a cichlid, *Cichlasoma nigrofasciatum*, were obtained by breeding these fish in thermostatically controlled aquaria.

Experiments were set up in finger bowls of 200-ml capacity, one and two liter finger bowls, one and two liter battery jars, or ten gallon aquaria. A control and a test group was used for each experiment. There were no copepods in control aquaria. Fry in test groups were subjected to cyclopoid concentrations equivalent to and ranging from 10 to 500 individuals per liter. Most of the experiments were run within the 20 to 100 copepods per liter range, which is the order of magnitude of the population density found in many freshwater lakes and ponds. Morgan (1958) in his extensive studies of the plankton of 11 one-acre ponds, however, found as many as 247.5 *C. bicuspidatus*, *Eucyclops agilis* and *Mesocyclops* sp. per liter in one of the ponds on April 30, 1955.

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A count of the fry in each aquarium was made daily. Direct observation of the test group was maintained much of the time to verify that death of the fry was actually due to cyclopoid attacks.

RESULTS

Mortality of all species one and two days after hatching was from 25 to 50 percent in the aquaria containing *C. vernalis*, *C. bicuspidatus* and *Mesocyclops leuckarti*. That in the control groups which contained no cyclopoids was from 1 to 5 percent. Table 1 represents a summary of 49 experiments showing the mortality of fry after three days with cyclopoid copepods. In all experiments grouped together, there was a distinct difference in survival between those containers having cyclops and those without. The 50 percent mortality in the sunfish control group probably indicates inadequate numbers of fry. Table 2

TABLE 1

A summary of 49 experiments showing mortality of various species of fry after three-days exposure to cyclopoid copepods in concentrations below 200 individuals per liter

Type of fry	Cyclops per liter	Number of fry	Mortality	% loss
Gourami	0	51	1	2.0
Gourami	6.6-125	81	25	30.9
Sunfish	0	14	7	50.0
Sunfish	25-150	28	24	85.7
Cichlid	0	73	13	17.8
Cichlid	3.2-186	124	96	77.4
Zebra	0	47	5	10.6
Zebra	10-169	44	33	75.0
Rock bass (?)	0	24	3	12.5
Rock bass (?)	20-50	21	6	28.6

TABLE 2

Summary of 24 experiments showing mortality of bluegill fry after five days exposure to various concentrations of Cyclops vernalis and Cyclops bicuspidatus

Cyclops per liter	Number of fry	Mortality	% loss
0	69	7	10.2
10-50	30	24	80.0
51-100	19	18	94.7
101-150	32	32	100.0

shows that as the number of cyclopoids increased, deaths of fry also increased over a period of five days. Four experiments were devised involving 27 control fry, 28 test fry and 200 or more cyclops per liter. Mortality in the test group was 46.4 percent, and no deaths in the control group after one day. By the second day there had been a 60.7 percent loss of test fry compared to a 7.5 percent loss in the control.

It was found that many of the fry that were 8 mm or more in length could avoid or survive damage from cyclopoids in concentrations equivalent to 1000 per liter. The size at which fry can withstand attacks probably varies with each species, activity or motility of the fry being perhaps of prime importance to survival.

The method by which a larval fish is destroyed by a cyclopoid is essentially that

described by Davis (1959). In many cases copepods were observed attached to and apparently chewing on the tail, head, body or yolk sac region of fry. If the yolk sac were the initial point of attack, it would usually rupture only a few minutes after the attachment of the copepod. Some of the yolk would flow out and coagulate. Death of the fry followed almost immediately. If the tail were attacked, mortality of the fry would appear to depend on the persistence of the cyclopoid and the amount of tissue devoured. If a cyclopoid attacked the head region, death of the fry was generally noted within two or three minutes.

CONCLUSIONS

These experiments indicate that mortality and predation of fry increase as the concentration of the cyclopoids increases. It is suggested from these experiments that certain cyclopoid copepods may be one biologic factor in reducing or lowering some fresh water fish populations. Overpopulation of fish would occur within a very short time if not for a multitude of physical, chemical and ecological factors.

ACKNOWLEDGMENTS

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I wish to extend my sincere thanks to Dr. C. A. Dambach, Director of the Natural Resources Institute; Dr. L. S. Putnam, Director of the F. T. Stone Laboratory; and Dr. T. M. McMillion, Geneva College, for the liberal use of materials and equipment. The assistance of Mr. Harold Wasko, Director of the Toledo Aquarium, in providing bluegill fry, and of Mr. Glenn Flittner, in providing sheephead fry, is gratefully acknowledged. Special thanks are due Dr. C. C. Davis who originally stimulated my interest in this problem and Dr. A. C. Broad for his many helpful suggestions.

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Geology of the Great Lakes. Jack L. Hough. University of Illinois Press. xviii+313 pp. \$8.50.

This is the first book on the subject since 1915, and the author presents much information discovered in the past 43 years. The book is organized logically into two main parts, with Part I presenting descriptions of the present lakes, good discussions of the limnologic character of the water and sediments, a preglacial and glacial history of the region, and a consideration of relative ages and the methods of age determination. Part II comprises a history of lake stages, with detailed records of each basin and summarizing with the radiocarbon chronology of Great Lakes history.

This well-organized book has 75 illustrations, most of them maps and sketches but there are a few photographs. The format is pleasing, the paper and printing is excellent, and the bibliography and index are adequate. The text is detailed enough to have value for scientists but the language makes it suitable for adult readers.

THOMAS H. LANGLOIS

FOUR NEW DOLICHOPODIDAE FROM THE
EASTERN UNITED STATES
(DIPTERA)

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A review of the known Dolichopodidae of the Eastern United States has revealed the following apparently undescribed species.

***Telmaturgus brevicornis* n. sp.**

Male.—Length, 2 mm; wing, 2 mm by 0.6 mm. Face very narrow below, becoming wider above, silvery white; front broad, slightly widened above, shining metallic violet; palpi and proboscis yellowish; antennae brownish-black; first segment short, only slightly wider apically, bare above; second segment short with a ring of fine setulae; third segment about as long as wide, bluntly pointed, with a dorsal arista; lower orbital cilia pale.

Mesonotum dark metallic bronze color with a median bluish streak and with black spots above the bases of the wings; pleurae a little darker with slight whitish pollen; acrostichal bristles very small, biseriata; dorsocentrals large, with four pairs; scutellum with two widely separated bristles and a few fine setulae; propleurae bare in the middle.

Coxae and the hind margin of the pleurae yellow; fore coxae with light brown bristles apically; middle and hind coxae with a black external bristle; middle coxae with black hairs on the anterior surface; fore and middle femora and tibiae yellow, hind femora yellow on the basal half; hind femora toward the apex, hind tibiae and tarsi, fore and middle tarsi toward the tips brownish; middle and hind femora with preapical bristles; middle tibiae with anterodorsal bristles near the basal fourth and distal third, a posterodorsal near the middle, and three apical bristles; hind tibiae with about six small bristles dorsally, approximately paired near the basal, middle and distal fourths, the middle pair longest; length of the fore tarsi as 12-6-4-3-3, middle tarsi as 17-9-6-5-4, hind tarsi as 11-14— with distal three joints broken off.

Wings narrowly oval, hyaline; with the third vein slightly bent backwards; fourth vein essentially straight beyond the crossvein; crossvein perpendicular to the last part of the fourth vein, one third as long as the last of the fifth vein. Calypters and halteres pale yellow; cilia of the former not observed.

Abdomen cylindrical, mostly dark bronze-colored with violet tinges; second and third segments yellowish, especially on the sides; hairs and bristles black; hypopygium small, imbedded in the tip, brownish-black; outer genital lamellae small, oval, brownish-black, with fine rather pale hairs.

Holotype male from herbaceous vegetation on stream bank, Duke Gardens, Durham, North Carolina, May 7, 1959, collected by Hazim Daoud. Type in the author's collection.

The genus *Telmaturgus* has been considered until now as monotypic and as being restricted to the palearctic region. However, in addition to the present species, *T. satrapa* (*Diaphorus satrapa* Wheeler) and a yet undescribed species from eastern Tennessee belong to this genus.

***Peloropecodes frater* n. sp.**

Male.—Length, 2 mm; wing, 2 mm by 1 mm. Face narrow, wider above, with transverse ridge near the mouth; front broad, narrowing below; face and front with yellowish-gray pollen nearly obscuring the dark ground color; palpi dark with coarse whitish pollen; proboscis dark brown; antennae dark brown, covered with fine pale hairs; first segment about as long as wide, bare above; second segment slightly shorter and wider, with a ring of setulae which are longest dorsally; third segment still wider at the base, a little longer than wide, lower margin slightly convex, upper margin nearly straight beyond the arista, rather pointed at the tip; arista dorsal near the basal third, about twice as long as the antennae, jointed at its basal third; lower orbital cilia except lowermost pair, with pale tips.

Mesonotum metallic dark green, dulled with yellow pollen, pollen whitish on the sides; acrostichals rather short, uniseriate, ending before the rather flattened area on the posterior slope; dorsocentrals larger, with six pairs; two large scutellar bristles with a pair of smaller hair-like bristles toward the outer side; a fine rather pale bristle above the fore coxae.

Fore coxae mostly brown, middle and hind coxae yellow with the outer surface brownish; fore and middle coxae with pale hairs anteriorly grading into dark bristles distally; middle and hind coxae with a black bristle on the outer surface; legs yellow with slight brownish color on the middle of the fore and upper edge of the hind femora, toward the tips of the fore and middle tarsi and broadly on the tips of the joints of the hind tarsi; middle and hind femora with distinct preapical bristles, hind femora with numerous long setulae near the preapical bristle; middle tibiae with two long anterodorsal bristles and two shorter posterodorsals paired near the basal fourth and middle, four apical bristles; hind tibiae with bristles short, two paired near the basal fourth, an anterodorsal just before the middle, a posterodorsal near the third fifth, three apicals; length of the fore tarsi as 17-7-5-4-5, middle tarsi as 24-15 11-7-6, hind tarsi as 13-19-14-9-7.

Wings oval, hyaline, with black veins; third and fourth veins parallel beyond the posterior crossvein, bent slightly backwards; crossvein perpendicular to the basal part of the fourth vein, about half as long as the last of the fifth vein; sixth vein represented by a slight fold. Calypters and halteres yellow with tips of the former and their cilia black.

Abdomen about as long as the thorax, metallic green dulled with yellow pollen on the first segment dorsally; hairs and marginal bristles black; hind margin of the fourth segment ventrally projecting with a slender rearward projection from the middle; hypopygium black; appendages yellow; outer appendages with a sharp bend followed by a slightly broadened portion appearing to have a hole in it, the tip with a very slender point; inner appendages and what appears to be the ventral sheath exerted as far as the outer appendages.

Holotype male on moist rock by stream, Duke Gardens, Durham, North Carolina, May 30, 1959. Type in the author's collection.

The new species superficially resembles *P. brevis* (Van Duzee) very closely, but in having the inner hypopygial appendages as long as the outer it approaches *P. bicolor* (Van Duzee). The short but ventrally convex third segment of the antennae is distinctive, and I have not seen a ventral marginal projection on an abdominal segment in any other *Peloropecodes*.

Condylostylus viridivittatus n. sp.

Male.—Length, 3 mm; wing, 3 mm by 1.2 mm. Face rather wide, metallic green, nearly obscured by white pollen on the lower two-thirds; front broad, shining metallic green, very slightly pollinose; occiput strongly concave on each side of the ocelli; palpi and proboscis translucent yellow, palpi with two black bristles and numerous shorter pale hairs; antennae yellow; first two segments nearly the same width, each a little shorter than wide, first bare above, second with a ring of setulae longer above and below; third segment very short and conical, covered with short white pubescence; arista dorsal and basal; eyes with very short pubescence in the lower part and along the hind margin; lower orbital cilia very fine and pale.

Mesonotum yellow with a longitudinal, median, metallic green stripe; disk of the scutellum metallic green with margins yellow; whitish pollen evident on the pleurae; three pairs of large acrostichal bristles; six pairs of dorsocentrals, anterior pair very small; scutellum with two strong bristles and two hairlike bristles; a slender pale bristle and a few pale hairs above the base of each fore coxa.

Coxae yellow; fore coxae with a crest of stout pale bristles on the outer edge near the base followed by numerous pale hairs, with a row of black hairs on the inner edge grading into a crest of black bristles at the tip; middle coxae with black hairs and distally black bristles; hind coxae with a few pale setulae and a strong black bristle on the outer surface; legs yellow with hairs and bristles black, denser toward the tips of the tarsi and almost obscuring the ground color; femora without true preapical bristles; fore tibiae with only a very short bristle dorsally near the basal fifth; middle tibiae with an anterodorsal bristle near the basal fourth, the two posterodorsals and a ventral bristle very short, three or four apical bristles; hind tibiae with a dorsal bristle near the basal fourth and with two apical bristles; hind basitarsi with two or three bristle-like hairs at the base ventrally which superficially appear to be apical bristles of the tibiae; distal joints of the tarsi flattened and slightly widened; length of the fore tarsi as 65-19-15-10-5, middle tarsi as 65-25-15-8-5, hind tarsi as 50-25-14-9-5.

Wings elongate oval, hyaline; second and third veins curved gradually backwards to near their tips which are widely separated; last section of the fourth vein about half as long as the basal part, slightly sinuous, ending before the margin; a fork from near the tip of the fourth

vein directed forward and slightly basally, with a rather sharp bend near the middle and straight and directed longitudinally in the last half, ending just before the apex of the wing near the tip of the third vein; posterior crossvein oblique with the anterior end more distal, half again as long as the last part of the fifth vein; sixth vein represented by a slight fold. Calypters and halteres yellow; cilia of the former black.

Abdomen mostly yellow; hairs and marginal bristles black; hind margin of the third segment and all of the fourth to the sixth segment with greenish and coppery tinges; hypopygium very small, dark; outer genital lamellae black, rather crescent-shaped, somewhat pointed apically, with a few black hairs.

Holotype male on foliage by shaded stream, Rock Creek Park near Silver Spring, Washington D. C., June 25, 1959. Type in the author's collection.

Of the characters used to distinguish *Condylostylus* from *Sciapus* I have found the color of the cilia of the calypters to be the most dependable. For this reason the present species has been considered to be a *Condylostylus* in spite of having only two strong scutellar bristles and a primarily yellow body color by which it resembles three neotropical species of *Sciapus*. The crest of pale bristles on the fore coxae appears to be a distinctive feature.

***Hercostomus brunneifacies* n. sp.**

Male.—Length, 2.5 mm; wing 2.5 mm by 1.1 mm. Face broad, slightly narrower below, with numerous short black bristles on the lower third, covered with brown pollen; front broad, dark metallic green with slight gray pollen; palpi and proboscis black; antennae black; first segment widened distally from a narrow base, with a few short hairs above; second segment about as long as the first but wider, longer on the sides, with a ring of setulae which are longer above and below; third segment about as wide as long, rather blunt, covered with short white pubescence, with a dorsal arista near the base; orbital cilia black.

Thorax dark metallic green with only slight yellowish pollen on the mesonotum, with thicker white pollen on the pleurae; fore part of the mesonotum with many hair-like bristles; acrostichals biserial, about a third as long as the dorsocentrals; six pairs of long dorsocentrals, the fifth pair more approximated; scutellum with two strong marginal bristles and with numerous fine setulae on the disk and margin; a black bristle above each fore coxa; very fine pale setulae before the posterior spiracle.

Coxae black, fore and middle coxae with black hairs and distally with bristles, middle and hind coxae with a strong black external bristle; femora, tips of fore tarsi, middle tarsi from the tip of the first joint, hind legs from the tips of the tibiae black; fore, middle and basal two thirds of hind tibiae yellow; middle and hind femora with a preapical oristle, middle femora also with a preapical-like bristle on the inner side; fore tibiae with only short hairlike bristles, only two of the anterodorsal row nearly as long as the width of the tibiae, these nearly paired with two short posterodorsals; middle tibiae with anterodorsal and posterodorsal bristles paired near the basal sixth and with a pair and a ventral bristle just beyond the middle, a third anterodorsal near the basal fourth, with five apical bristles; hind tibiae with anterodorsal and posterodorsal bristles nearly paired at the basal sixth and third, an anterodorsal near the distal third, a posterodorsal near the middle, with two small ventral bristles, two apicals and one subapical; length of the fore tarsi as 9-4-3-2-2, middle tarsi as 12-6-5-3-3, hind tarsi as 8-10-6-5-4.

Wings oval, very slightly brownish tinged; third and fourth veins nearly straight and parallel beyond the posterior crossvein, fourth ending distinctly before the tip of the wing; crossvein perpendicular to the base of the fourth vein, about two thirds as long as the last part of the fifth vein; sixth vein represented by a slight fold. Calypters and halteres yellow; cilia of the former black.

Abdomen a little longer than the thorax, metallic dark green, with the hairs and the slightly longer marginal bristles black; hypopygium black; outer genital lamellae somewhat wider than long, rather truncate apically, brown with black cilia; inner appendages yellow.

Holotype male on foliage along a marshy stream, Bronx Park, Bronx, New York, June 26, 1959. Type in the author's collection.

Of the group of species with short bristles on the face of the male no other has the face covered with brown pollen. The only other species of the group having short and truncate hypopygial lamellae has the hind tibiae mostly black.

DEMONSTRATION OF THE CHEMOTROPISM OF POLLEN TUBES IN VITRO IN FOUR PLANT SPECIES¹

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The phenomenon of chemotropism has been reported for pollen tubes (Beck and Joly, 1941; Brink, 1924; Lidforss, 1899; Miyoshi, 1894a; Molisch, 1893; and Tsao, 1949) for the spores of certain fungi (Graves, 1916; Miyoshi, 1894b) and for the organs of higher plants such as roots (Newcombe and Rhodes, 1904). Although numerous species of higher vascular plants have been studied, only about 10 species produced pollen which exhibited chemotropic characteristics toward some part of the pistil of the same species. Examples of this phenomenon were first reported at the end of the 19th century (Lidforss, 1899; Molisch, 1893). The literature up to 1924 has been reviewed by Brink. Tsao conducted an extensive study of the chemotropism of pollen tubes and reported her results in 1949 with a review of the literature up to that time. The chemical or chemicals responsible for the tropic response have never been isolated.

This present study was initiated as a survey of a small number of species of plants, most of which had never been studied previously, for the possibility of the occurrence of the chemotropic characteristic of the pollen tubes toward plant parts of the same species.

MATERIALS AND METHODS

In most of the research reported in the literature, a method *in vitro* was used to demonstrate chemotropism of pollen tubes. This usually consisted of placing a small segment of some part of the pistil or other floral part on a semisolid medium and then placing the pollen around this tissue.

The method of Tsao (1949) was used in this study with only minor modifications. Pollen was collected from plants grown in the greenhouse. Where possible, flowers just at anthesis or a day or so past anthesis were used in these studies. However, no attempt was made to use pollen of exactly the same age in the tests. Flowers were brought intact into the laboratory and the pistil, other floral organs, or plant parts were cut into small segments. The size of these segments varied, but for segments of the ovary, for the petals and sepals, pieces about 3-5 mm square were used. The style provided small cylinders about 3 mm in length. Ovules, when used in these studies, were excised intact from the ovary. The method consisted of placing about five segments from each floral organ at approximately equally spaced intervals on a semisolid medium in petri dishes. Pollen from the same species was then placed around the floral tissue segment at a distance of about 1 mm with a dissecting needle, under a binocular microscope. Thus, each test or treatment consisted of at least five duplicates in any one experiment and each experiment was repeated at least once. Positive chemotropism in a given species was arbitrarily considered as the growth of about 90 percent or more of the pollen tubes from grains placed 1 mm from the test segment toward that segment.

The medium consisted of 1 percent agar-agar, 10 percent sucrose, and 100 ppm Difco Yeast extract. The mixture was autoclaved for 15 minutes at 15 pounds pressure. Before the medium had cooled completely it was poured into

¹Contribution from the Department of Botany and Plant Pathology, The Ohio State University, paper No. 542.

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petri dishes to a depth of about 4 mm. The plant materials were placed on the medium after it had cooled and the dishes were examined under low power at intervals beginning one hour after the pollen was placed on the medium. For species whose pollen did not germinate well, the dishes were examined the following day and then discarded. For species whose pollen germinated readily on the medium employed here, the results could usually be read in from two to four hours. No attempt was made to adjust the medium in order to obtain good growth of the pollen of all the species studied.

RESULTS

Ten species of plants, representing ten genera and seven families were studied. The results are shown in table 1. Positive chemotropism was demonstrated by pollen of one species of *Aloe*, one species of *Clivia*, one species of *Gasteria*, and one species of *Haworthia*, to segments of one or more floral organs of the same species

TABLE 1

The results of the study of the effect of one or more floral parts on the direction of growth of pollen of the same species in vitro

Plant	Family	Positive chemotropism
<i>Aloe confusa</i>	Liliaceae	+ ¹
<i>Antirrhinum majus</i>	Scrophulariaceae	— ²
<i>Clivia nobilis</i>	Amaryllidaceae	+
<i>Crinum</i> sp.	Amaryllidaceae	—
<i>Epiphyllum</i> sp.	Cactaceae	—
<i>Gasteria verrucosa</i>	Liliaceae	+
<i>Haworthia planifolia</i>	Liliaceae	+
<i>Impatiens sultani</i>	Balsaminaceae	—
<i>Kalanchoe</i> sp.	Crassulaceae	—
<i>Nicotiana</i> sp.	Solanaceae	—

¹90% or more of the pollen tubes grew toward the floral part embedded in the medium.

²Growth of the pollen tubes was at random.

TABLE 2

Plant species in which pollen tube chemotropism was demonstrated

Plant	Tissues toward which pollen tubes of the same species grew
<i>Aloe confusa</i>	Stigma, style, petal, sepal
<i>Clivia nobilis</i>	Stigma, style, ovulary wall, ovule, leaf, petal, sepal
<i>Gasteria verrucosa</i>	Stigma, ovulary
<i>Haworthia planifolia</i>	Stigma, style, ovulary

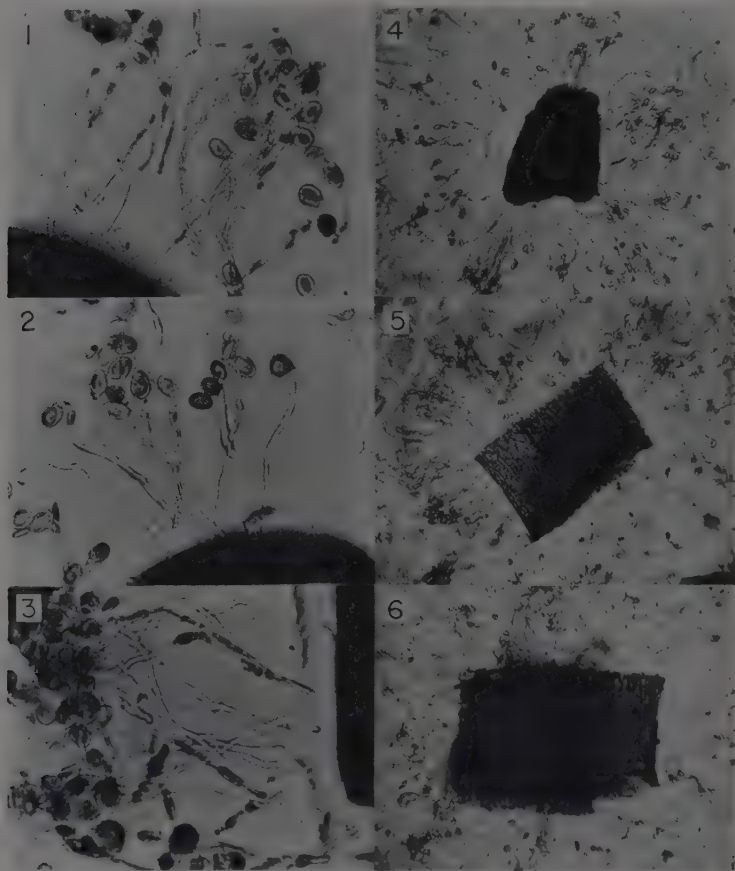
respectively. The pollen of six other species grew at random when placed around segments of floral organs of the same species.

In addition to parts of the ovulary such as the stigma, style, ovulary wall, and ovule, other floral parts such as petals and sepals, as well as leaf tissue for certain species, elicited the chemotropic response to pollen of the same species. These results are given in table 2.

Photomicrographs were made of the chemotropic response of the *Clivia nobilis* pollen to parts of the flower and to leaf segments (fig. 1-6).

Additional studies were carried out on the pollen of *Clivia nobilis* to learn more of the nature of the chemotropic factor.

With this species it was possible to confirm the results of Tsao (1949) that the chemotropic factor is heat stable and readily diffusable into agar. Even after heating segments of the ovulary in water at 212° F for 15 minutes, it was possible to obtain the tropic response to this tissue equivalent to the unheated controls. When ovules (6-12) were placed on agar cubes (about 4 mm³ in volume) and allowed to stand for 5-8 hours, it was possible to obtain the chemotropic response of pollen of *Clivia nobilis* to these cubes *after* the ovules had been removed (fig. 7). Again the response was approximately equivalent to controls using excised intact ovules on agar plates as previously described. The pollen was placed approxi-



FIGURES 1-6. The demonstration of the growth of pollen tubes of *Clivia nobilis* toward vegetative and floral parts of the same species. FIGURES 1-3. Growth of the pollen tubes toward a segment of the style; toward an entire, excised ovule; and toward a segment of a leaf, respectively. FIGURES 4-6. Growth of the pollen tubes toward the stigma, toward a segment of the sepal, and toward a segment of the petal, respectively.

mately 1 mm from the agar cubes as in the other experiments with tissue segments. In order to test the possibility that pollen tubes of *Clivia nobilis* would grow toward any object embedded in the agar, experiments similar to those described were carried out with the tissue segments replaced by clean glass beads. Growth of the tubes of *Clivia nobilis* pollen grew completely at random around the glass beads.



FIGURE 7. The demonstration of the growth of pollen tubes of *Clivia nobilis* toward an agar cube containing a diffusate from ovules of the same species.

DISCUSSION OF RESULTS

The data presented here in general corroborate the results of Tsao (1949). However, it was not possible to demonstrate chemotropism in the unidentified variety of *Antirrhinum majus* studied here. The active factor for *Clivia nobilis* may be chemically similar to the unknown chemical reported by Tsao (1949) for *Lilium superbum* since both are heat stable, water soluble, and diffusable into agar.

Tsao (1949) reported that the petals, sepals and leaves of *Antirrhinum majus* did not attract the pollen tubes of the same species. In the present study it was found that the sepal, petal, and leaf segments of *Clivia nobilis* did attract the pollen tubes of the same species. Thus, it appears that at least in this species the active factor is distributed throughout the above ground parts of the plant. It was also possible to demonstrate chemotropism of the pollen tubes of *Aloe confusa* to segments of the petal and sepal in addition to the tropic response to the stigma and to segments of the style (table 2).

SUMMARY

1. Of ten species of plants investigated, it was possible to demonstrate the chemotropism of the pollen tubes of four species to tissue segments of the same species. These four species are *Aloe confusa*, *Clivia nobilis*, *Gasteria verrucosa*, and *Haworthia planifolia*.

2. The pollen of these four species exhibited the chemotropic response to various segments of the pistils of the respective species, and in addition the pollen of *Aloe confusa* and *Clivia nobilis* grew toward petal and sepal segments. The pollen of *Clivia nobilis* also demonstrated the chemotropic response to leaf tissue segments.

3. The active factor bringing about the chemotropic growth of the pollen tubes of *Clivia nobilis* was found to be heat stable, water soluble, and diffusible into agar.

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THE BRYOPHYTE FLORA OF CLINTON COUNTY, OHIO¹

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Clinton County is an agricultural county in southwestern Ohio from which 13 species of bryophytes were reported prior to this study. The 400 square miles of the county are covered by glacial soil deposited by three Pleistocene ice masses. The county has a variety of microhabitats from which 105 mosses and 9 hepatics are listed. Two of the mosses, *Bryum pendulum* (Hornsch.) Schimp. and *Weissia microstoma* (Hedw.) C. Muell., were previously unknown from Ohio. Several species represent important second finds in the state. Although the bryophyte flora is essentially that of the great lakes states, several tropical and subtropical species are present in the flora.

ANNOTATED LIST OF SPECIES

* previously recorded for Clinton County

† new record for Ohio

Abbreviations preceding the collection numbers indicate the townships, as follows:

A	Adams	G	Green	M	Marion	WA	Washington
CH	Chester	J	Jefferson	U	Union	WI	Wilson
CL	Clark	L	Liberty	V	Vernon		

¹Part of a thesis submitted in partial fulfillment of the requirements for the degree of Master of Arts in Teaching.

²Present address: 1923 East 8th St., Davis, California.

All specimens cited are deposited in the Miami University Herbarium. Specimens deposited in The Ohio State University Herbarium are designated OS; those from the University of Cincinnati CINC; those from Baldwin Wallace College BW.

Musci

- Atrichum angustatum* (Brid.) B.S.G. A, 302; CL, 42; V, 274. On soil in exposed or unexposed places, usually in damp woods.
- Atrichum macmillani* (Holz.) Frye. A, 285; U, 62. On sandstone.
- **Atrichum undulatum* (Hedw.) Beauv. A, 296, 303; CL, 17, 83; J, 465, 468, 471; U, 23, 113; V, 200A, 203, 239; WA, 67. Green Township, J. S. VanDervoort, 1892 (OS). Very common on soil and humus in woods.
- Polytrichum commune* Hedw. V, 240. On soil in moist woods.
- Polytrichum ohioense* Ren. and Card. V, 187, 192, 228, 259. On damp soil in woods.
- Fissidens bryoides* Hedw. V, 188. On shaded soil.
- Fissidens cristatus* Wils. A, 307, 339. On tree bases.
- Fissidens debilis* Schwaegr. V, 273. On a submerged rock in a running stream in a woods.
- Fissidens obtusifolius* Wils. A, 207; L, 367, 368, 370. On moist limestone and sandstone, occasionally on bark at base of trees.
- Fissidens osmundioides* Hedw. V, 218. On moist shaded soil.
- Fissidens taxifolius* Hedw. A, 338; J, 427, 460, 470, 477; V, 164, 165, 208, 214, 215, 217, 264; WA, 10. Very common on damp shaded soil. According to Robinson (1957) *F. taxifolius* is less common than *F. osmundioides* in southeastern Ohio.
- **Ceratodon purpureus* (Hedw.) Brid. A, 294; J, 442. Reported by Giesy (1957) from University of Cincinnati specimen. Not seen. Number 294 on bare and compact clay soil; 442 on top of a moist and shaded tree stump.
- **Ditrichum pallidum* (Hedw.) Hampe. U, 106; V, 132, 168; WA, 6. Jefferson Township, Margaret Fulford, 1930 (CINC). On bare soil, wood and especially concrete bridge abutments where damp.
- Ditrichum pusillum* (Hedw.) E. G. Britton. A, 337; CH, 397; V, 221. On exposed clay soil.
- **Pleuroidium subulatum* (Hedw.) Lindb. Reported by Giesy (1957) from University of Cincinnati specimen. Not seen.
- Dicranella heteromalla* (Hedw.) Schimp. CL, 21; V, 153. On moist clay bank and on bark at base of tree.
- Dicranella varia* (Hedw.) Schimp. A, 330; J, 481; U, 38; WA, 30. On shady sandy and clay soils. Number 30 confirmed by Dr. Lewis E. Anderson.
- Dicranum condensatum* Hedw. V, 198. On rotting log.
- Dicranum fulvum* Hook. A, 324; V, 231, 278. Found exclusively on bark. Robinson (1957) reports it from southeastern Ohio on shaded rock.
- **Dicranum scoparium* Hedw. Jefferson Township, Margaret Fulford, 1930 (CINC). Growing in a mat around a pin oak tree in a woods near Westboro.
- Dicranum viride* (Sull. & Lesq.) Lindb. A, 290; V, 195. On tree bases and soil around the base of trees.
- **Leucobryum glaucum* (Hedw.) Schimp. Jefferson Township, Margaret Fulford, 1930 (CINC). On the soil in an oak-beech woods.
- Barbula fallax* Hedw. A, 325; CL, 36B. On soil of open areas.
- Barbula unguiculata* Hedw. A, 301, 304; CH, 413; U, 423; V, 124, 131, 134. On rock, particularly concrete bridge abutments, bare soil, and occasionally on bark. Not uncommon.
- Gymnostomum calcareum* Nees & Hornsch. CH, 419; V, 154. On moist calcareous rocks.
- Gymnostomum recurvirostrum* Hedw. U, 35; V, 141; WA, 63. On rock. Found once growing on a stump in a swampy area at the northeastern edge of Lake Cowan.
- Tortella humilis* (Hedw.) Jennings. J, 462; V, 150; WA, 15. On concrete abutments, roots of trees in woods.
- Tortella tortuosa* (Turn.) Limpr. V, 127. On limestone.
- Tortula mucronifolia* Schwaegr. WA, 28. On concrete bridge abutment.

- † *Weissia microstoma* (Hedw.) C. Mull. A, 334. On clay soil. This is the first authenticated specimen from Ohio. (Andrews, 1924).
- Weissia viridula* Hedw. CH, 378; U, 95; V, 140, 161. Usually on bare soil among grasses.
- Grimmia apocarpa* Hedw. U, 98; V, 155. On exposed rock and rotting log.
- Aphanorhegma serratum* (Hook. & Wils.) Sull. U, 71. Found growing over several square feet in our vegetable garden in Wilmington. Probably common.
- Funaria flavicans* Mx. U, 48, 94, 492. On bare soil, gravel driveway, floor of greenhouse.
- Funaria hygrometrica* Hedw. CH, 378; U, 493. Number 378 found on soil with *Weissia viridula*; number 493 on dirt floor of greenhouse.
- * *Physcomitrium turbinatum* (Mx.) Brid. CL, 39, 424; V, 76; WI, 360. Jefferson Township, Margaret Fulford, 1930 (CINC). On bare soil, usually in open fields.
- Orthotrichum garrettii* Grout & Flowers. J, 430. On a dry granite boulder used in building the dam at Westboro. Recorded previously only from Franklin County.
- Orthotrichum obtusifolium* Brid. A, 281; J, 450. On bark of trees.
- Orthotrichum ohioense* Sull. & Lesq. A, 309. On tree bark.
- Orthotrichum pumilum* Dicks. A, 88, 288; CH, 398; G, 343. On trees.
- Orthotrichum pusillum* Mitt. A, 333A; J, 433; M, 488. Loose tufts on bark of trees.
- Orthotrichum strangulatum* Schwaegr. A, 321, 326; CH, 400, 408, 412, 415; J, 429, 431, 438, 456; M, 490; U, 16; V, 19, 123, 125, 146, 151, 160, 163, 258; WA, 14, 29. Very common on concrete abutments and limestone and granite rocks.
- Orthotrichum sordidum* Sull. & Lesq. CH, 384. On tree bark. Giesy (1957) cites one specimen from Lesquereux. No data given.
- Aulacomnium heterostichum* (Hedw.) B. S. G. V, 208, 256, 263. On rich soil and humus in woodlands.
- Philonotis fontana* (Hedw.) Brid. V, 72. Found on a tree stump in the swamps at the north-eastern edge of Lake Cowan.
- Bryum argenteum* Hedw. A, 313; CH, 407B; J, 425; U, 24, 93; V, 72; WA, 4, 5. On soil, rocks, in dry situations. Very common along sidewalks, paths, railroad tracks and ash piles.
- Bryum bicolor* Dicks. A, 326; CH, 407A. On sandy soil in moist situations. Once collected with *B. argenteum*. Previously recorded only in Athens County.
- * *Bryum caespitium* Hedw. CL, 43; U, 96, 495; V, 139, 280. Jefferson Township, Margaret Fulford, 1930 (CINC). Usually on soil low in organic matter, occasionally on rock.
- Bryum capillare* Hedw. WA, 64. On tree stump in swampy area at northeastern edge of Lake Cowan.
- Bryum cuspidatum* (B. S. G.) Schimp. A, 305; WA, 5. On soil and rock.
- † *Bryum pendulum* (Hornsch.) Schimp. V, 122. Collected on a limestone rock in the creek bed near Springhill Road, Clarksville. This species has not previously been reported in Ohio. Common farther north and reported to be rather common in Indiana (Welch, 1957).
- * *Pohlia nutans* (Hedw.) Lindb. A, 311, 314, 317, 319; J, 443; V, 162, 254, 262; WA, 68. Jefferson Township, Margaret Fulford, 1930 (CINC). On soil, humus, rocks, in various situations.
- Pohlia wahlenbergii* (Web. & Mohr.) Andrews. A, 313. On wet clay bank.
- Rhodobryum roseum* (B. S. G.) Limpr. A, 283. On moist soil. Not common.
- Mnium affine* Bland. CH, 411; CL, 84; U, 57; V, 156, 166, 181, 200A, 276; WI, 359. On soil and rotting logs.
- * *Mnium cuspidatum* Hedw. A, 322, 335; CH, 389, 418; CL, 34; J, 444, 463, 466, 467, 469; M, 484; U, 55, 62, 102, 104A, 107; V, 20, 74, 79, 118, 120, 133, 147, 176, 179, 205, 208, 245, 261, 269; WA, 26; WI, 357. Jefferson Township, Margaret Fulford, 1930 (CINC). Very common on rotting logs, on soil and humus in woods. *Lophocolea heterophylla* is often found growing with this species.
- Mnium punctatum* Hedw. A, 305, 312; V, 185; WA, 13. On rocks, logs, soil in moist situations.
- Amblystegium juratzkanum* Schimp. V, 238A, 247. On moist soil.
- Amblystegium serpens* (Hedw.) B. S. G. U, 49, 50, 51; V, 224. On shaded sandstone soil.
- Amblystegium varium* (Hedw.) Lindb. A, 332; CH, 388, 402; J, 474; U, 40; V, 126, 157; WA, 32; WI, 357. On concrete bridge abutments, moist limestone ledges, rotting wood and bark of trees. Common.

- Brachythecium acutum* (Mitt.) Sull. V, 115; WA, 70. On moist soil and humus.
- Brachythecium campestre* B. S. G. U, 58. On shaded soil.
- Brachythecium flagellare* (Hedw.) Jennings. CH, 380, 406; G, 342; V, 144, 171, 174, 208, 277; WI, 353. On soil, rotting logs and rocks near streams. Common.
- Brachythecium flexicaule* Ren. & Card. J, 479. On soil on moist bank.
- Brachythecium oxycladon* (Brid.) Jaeger. & Sauerb. A, 308; CH, 420, 421, 422; CL, 22; U, 112; V, 206. Base of trees, soil.
- Brachythecium rivulare* B.S.G. V, 189. In wet area on soil.
- Brachythecium rutabulum* (Hedw.) B.S.G. A, 91B; CL, 33, 44. On soil, humus and tree stumps.
- Brachythecium salebrosum* (Web. & Mohr.) B.S.G. A, 90; V, 183, 223; WA, 3, 25. Base of trees, soil.
- Campylium chrysophyllum* (Brid.) Bryhn. V, 167, 172. Roots of trees.
- Campylium stellatum* (Hedw.) Lange and C. Jens. CH, 395. On twig from tree.
- Cirriphyllum boscii* (Schwaegr.) Grout. A, 336; J, 482; U, 41; V, 170, 175, 209, 238B, 253. Usually on soil. Common.
- **Climacium americanum* Brid. Green Township, J. S. VanDervoort, 1892 (OS).
- Entodon cladorrhizans* (Hedw.) C. Mull. A, 286B, 340; U, 60; V, 77, 197. Usually on fallen trees, occasionally on soil, in shaded areas.
- Entodon seductrix* (Hedw.) C. Mull. A, 293; CH, 373, 375, 381, 382; J, 435, 455; U, 41, 59, 104B, 109; V, 8, 80, 225; WA, 7; WI, 363. Base of trees, concrete bridge abutments, soil, often in dry situations. Common.
- Eurhynchium hians* (Hedw.) Jaeger & Sauerb. A, 89, 91A, 92, 320; CH, 379, 391; V, 214. On soil.
- Eurhynchium serrulatum* (Hedw.) Kindb. A, 299; CH, 387, 396; CL, 45; U, 53, 111; V, 117, 119, 149, 205, 212, 257, 271; WA, 9, 69. Very common on tree bases, rocks and soil in woods.
- Eurhynchium strigosum* (Hoffm.) B.S.G. A, 329; CH, 414; G, 341; V, 190. On soil and tree bases.
- Heterophyllum halaanianum* (Grev.) Kind. CL, 37. On rotting wood.
- Homalotheiella subcapillata* (Hedw.) Card. V, 129. This specimen was collected on a limestone rock in shaded area.
- Hygroamblystegium irriguum* (Wils.) Loeske. A, 323; CH, 377, 386, 393, 404; U, 105; V, 169, 210, 226, 243. On rocks in streams, on soil at edge of streams and wet areas around trees. Common.
- Hypnum cupressiforme* Hedw. CH, 392, 410; J, 453; V, 78, 81, 229, 248; WI, 352. On soil, bark of dead trees.
- **Hypnum curvifolium* Hedw. Reported (Giesy, 1957) to be in the Baldwin Wallace College Herbarium. Not seen.
- Hypnum imponens* Hedw. V, 200B, 201, 244. On rotting wood.
- Hypnum reptile* Mx. V, 180. On soil in woods.
- Leptodictyum riparium* (Hedw.) Warnst. U, 52; V, 138; WA, 66. On rotten wood, tree stumps, in swamps; one specimen was found on wet limestone drippings.
- Plagiothecium denticulatum* (Hedw.) B.S.G. V, 74. On bark of dead tree with *Mnium cuspidatum*.
- Plagiothecium deplanatum* (Sull.) Grout. A, 295; U, 61. On soil.
- Plagiothecium muellarianum* Schimp. V, 136, 235, 268. On shady wet limestone cliff and shady soil in wooded ravine.
- Plagiothecium turfaceum* (Lindb.) Lindb. V, 177. On wood and humus in shaded moist woods.
- Platygyrium repens* (Brid.) B.S.G. A, 292, 297B; CH, 372, 376, 385; CL, 18; G, 349; U, 108; V, 75, 82, 202, 220, 241; WI, 354, 356. Very common on the bark of trees, logs and stumps.
- Pylaisia selwynii* Kindb. U, 100. Bark of living tree. Probably not uncommon.
- Anomodon attenuatus* (Hedw.) Huben. A, 282, 298, 306, 331; CH, 390, 394, 399; J, 448, 458; U, 99; V, 143, 152, 216, 237; WA, 1; WI, 365. On tree bases, soil and occasionally on rocks in shaded areas. Common.

- Anomodon minor* (Beauv.) Lindb. J, 475; V, 121, 142; WA, 2. Soil, limestone bluffs, tree trunks.
- Anomodon rostratus* (Hedw.) Schimp. V, 114, 133, 196, 199, 255, 279. On soil in wooded areas, logs, base of trees and on rocks in moist situations. Common.
- Leskea gracilescens* Hedw. A, 297A; CH, 416; CL, 46; G, 347; J, 432, 434, 436, 446, 464; U, 54, 97; V, 12, 31, 128, 147, 158, 186, 251; WA, 11; WI, 361, 366. On living trees, especially near the base; occasionally on soil. Very common. Robinson (1957) reports it common in southeastern Ohio on stones and cement culverts.
- Leskea nervosa* (Schwaegr.) Mylin. V, 73; WI, 358. Number 73 was found on the base of a tree; number 358 was growing on a brick in a woods. The only other record of this species for Ohio is from Ottawa County. It is uncommon this far south, and rarely found on stone.
- Leskea obscura* Hedw. A, 315. On bark of living tree, near the base.
- Leskea polycarpa* Hedw. A, 87, 88. On bark of living trees, near base.
- Thuidium delicatulum* (Hedw.) Mitt. CH, 403; V, 204. On soil in woods.
- Thuidium microphyllum* (Hedw.) Best. V, 145. On soil.
- Thuidium minutulum* (Hedw.) B.S.G. V, 182. On rotting wood.
- **Thuidium recognitum* (Hedw.) Lindb. Green Township, J. S. VanDervoort, 1892 (OS).
- Thuidium virginianum* (Brid.) Lindb. A, 289. On shaded soil.
- Leucodon julaceus* (Hedw.) Sull. A, 291. On bark of fallen trees.
- Clasmatodon parvulus* (Hampe) Sull. CH, 417; V, 219. On bark of living trees.

Hepaticae

- Phaeoceros laevis* (L.) Prosk. V, 227. Collected from tracks made by vehicles in moist clay soil. This liverwort has the reputation of being very rare (Steere, 1940). Hall (1958) records it as common in Athens County.
- Calypogeia trichomanis* (L.) Corda. V, 193. On moist soil growing with *Lophocolea heterophylla*.
- Chiloscyphus pallescens* (Ehrh.) Dumort. V, 194. Growing on wood with *Lophocolea heterophylla*.
- Conocephalum conicum* (L.) Dumort. U, 27; V, 267. Specimen number 267 was found on soil near Austin's woods; number 27 was on the soil in a potted ivy plant. This liverwort is known to be a common greenhouse weed.
- Frullania eborensis* Gottsche. A, 287, 310, 333B; CH, 402; CL, 36A; J, 449, 472; M, 489; V, 230, 232, 246; WI, 355. Very common on bark of living trees.
- Geocalyx graveolens* (Schrader.) Nees. A, 324; V, 222. On rotting wood.
- Solenostoma pumila* (With.) Steph. V, 266. Wet soil in Austin's woods near Lake Cowan.
- Lophocolea heterophylla* (Schrader.) Dumort. A, 286A, 316, 339; J, 466, 469; V, 170, 173, 174, 213, 233, 252, 272. On rotting logs, tree bases, soil. Very common.
- Porella platyphylloidea* (Schwein.) Lindb. A, 300, 318; CH, 374, 401; J, 454, 461; U, 110; V, 135, 148, 184, 234, 260; WI, 362. Very common on tree bases and moist soil.

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A PROPOSED ROLE FOR HYDROGEN PEROXIDE IN CARCINOGENESIS†

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According to a recent proposal (Kovacic, 1959), cancer may originate in the action of an agent, derived from either an artificial carcinogen or a spontaneous process, which directly or indirectly generates hydrogen peroxide in abnormally high concentration. Since this concept is applicable to a wide variety of carcinogens, it presents a broadly unified picture of the initiation phase of carcinogenesis. Plausible routes leading to high levels of peroxide were discussed in the preceding publication. The present report is concerned with the possible mode of action of hydrogen peroxide in its assigned role as the actual carcinogenic agent in the cases previously described.

There are three principal lines of evidence which buttress the proposition that hydrogen peroxide plays a key role in at least certain types of carcinogenesis.

1. Glass and Plaine (1953) made the very significant discovery that hydrogen peroxide produces melanotic tumors in *Drosophila melanogaster* (Plaine and Glass, 1955; Plaine, 1955a, 1955b).

2. Hydrogen peroxide is reported to be anti-carcinogenic (Hollcroft and Lorenz, 1952; Makino and Tanaka, 1955; Worrall, 1956; Holman, 1957; Sugiura, 1958).

3. Hydrogen peroxide possesses mutagenic properties (Demerec, 1953; Plaine, 1955a; for other references see Schumb, Satterfield and Wentworth, 1955a).

Since many carcinogens, such as ionizing radiation, nitrogen mustards and epoxides, possess mutagenic and cancer-inhibiting properties, these same characteristics in the case of hydrogen peroxide can be taken as additional evidence to strengthen the hypothesis.

It is worthy of mention that inorganic and organic hypochlorites, which are chemically similar to hydrogen peroxide, both enhance and inhibit tumor growth (Laszlo, Burk and Wight, 1959). The free radical decomposition of hypochlorites is discussed by Walling (1957).

In the present context, it is reasonable to assume that the cancer cell properties of low catalase content (Greenstein, 1954a) and high peroxide concentration (Rondoni and Cudkowicz, 1953) represent a continuation of the conditions which are critical for establishment of the malignancy.

With these concepts in mind, let us now consider the possible action of hydrogen peroxide in its effecting the transformation of a normal cell to a cancerous one, once the peroxide is present in abnormally high concentration. Existing evidence will be presented to support the contention that hydrogen peroxide, in the form of derived radicals, induces cancer by forming a template of simpler structure.

Cell Morphology and Histology

The idea of a common, or quite similar, mode of origin (Haddow, 1946-1947; Walpole et al., 1954) is consistent with the rather close resemblance between cancer cells regardless of the method of their generation. As Greenstein (1954b) pointed out "... tumors resemble each other more than they do normal tissues, or more than normal tissues resemble each other. . . ." A large number of investigators have noted that a wide variety of carcinogens induces a similar sequence of histological changes during the conversion of a normal cell to a malignant one

†This paper was presented in part at the Ohio Academy of Science meeting in Columbus, Ohio, on April 17, 1959.

(Rhoads, 1946; Farber, 1956; Haddow, 1957; Korpásky, 1959). Furthermore, Duryee et al., (1960) remarked on the striking similarity between human and amphibian neoplasms.

Energy Considerations

The evidence indicates quite conclusively that the cancer cell is at a lower, more favorable energy level than the normal cell. For one thing, it has a simpler, more primitive structure; i.e., the tumor cell is less differentiated. Also, the rate of growth is faster; it can grow at the expense of the normal cell. It is clear, however, that the normal cell does not revert spontaneously to the more favorable energy level -- apparently an energy barrier must be overcome (Schrödinger, 1944; Haddow, 1946-1947). Hydrogen peroxide possesses a high energy content (Schumb, Satterfield and Wentworth, 1955b), probably sufficient to provide the necessary energy of activation.

The foregoing considerations point to a gain in entropy when a normal cell is converted to a cancerous one (Rondoni, 1955; Flory, 1953). A more favorable entropy level for the template might be achieved in various ways, all of which would result in a simplification of structure: depolymerization; reduction in the degree of cross-linking; decrease in the number of strands comprising the helices; reduction in inter-chain forces, e.g., hydrogen bonding; or selective destruction of those nucleic acids concerned with specialized functions.

Role of Nucleic Acid

There is widespread agreement that nucleoproteins play an important role in the hereditary process, as well as in general cellular organization and function. Therefore, in connection with the hypothesis of template simplification, it is logical to focus primary attention on these vital cellular constituents. A number of investigators (e.g., Haddow, 1951; Leván, 1956; Hellström, 1959) have observed changes in chromosomal structure on application of carcinogens or in the transformation of normal to cancerous tissue. Recently, infectious DNA (deoxyribonucleic acid) was isolated (DiMayorca et al., 1959) from polyoma virus derived from leukemic mice (Stewart, Eddy and Borgese, 1958) and shown to be cancer inducing. Previously, Hays et al., (1957) and also Latarjet et al., (1958) reported cancer induction on injection of nucleic acid obtained from leukemic mice. Although the precise structural changes have not yet been elucidated, the evidence indicates that the DNA's from normal cells and from the corresponding cancer cells differ in certain of their properties (Polli and Semenza, 1955a, 1955b, 1956; Villa et al., 1955; Polli, 1957; Polli and Shooter, 1958; Polli et al., 1959; Sloan-Kettering Report, 1957-1959; DiMayorca et al., 1960). It might well be that a relatively small alternation in the DNA "... would lead to enormous differences in the functional elements synthesized at this locus" (Greenstein, 1954c).

Conflicting reports exist in the literature concerning the effect of hydrogen peroxide on nucleic acid. In some cases a marked depolymerization was noted, whereas in others there was essentially no change (Taylor, Greenstein and Hollaender, 1948; Krejci, Sweeny and Hambleton, 1949; Butler and Conway, 1950; Conway and Butler, 1952; Yamafuji, Hiramaya and Miyata, 1956). Butler and Conway (1950) suggested that the difference in behavior may be due to the presence in the sensitive samples of catalysts capable of converting hydrogen peroxide to active radicals.

Various workers (Scholes, Stein and Weiss, 1949; Butler and Smith, 1950; Butler and Conway, 1950) have demonstrated that the degrading action of hydrogen peroxide is greatly enhanced by the addition of ferrous ions, even for preparations of nucleic acid insensitive to hydrogen peroxide alone (Conway and Butler, 1952). Many investigators (e.g., Sparrow and Rosenfeld, 1946; Taylor, Greenstein and Hollaender, 1948; Scholes, Stein and Weiss, 1949) have reported the degrading action of ionizing radiation on nucleic acid. It is interest-

ing that the decrease in viscosity continues even after termination of irradiation (Taylor, Greenstein and Hollaender, 1948). The degradation has been attributed (Weiss, 1952; Conway, 1954) to attack by radicals derived from peroxides, hydrolysis of labile phosphate esters, and loss of hydrogen bonding. In 1950 Dickey advanced the hypothesis that the fundamental biological effect of irradiation is a free-radical promoted, depolymerization of deoxyribosenucleic acid. Similarly, drastic reductions in the viscosity of DNA mixed with hydrogen peroxide were obtained on short exposure to ultraviolet light (Butler and Smith, 1950; Smith and Butler, 1951). With irradiation alone, depolymerization also occurs, but the rate is considerably less than that in the presence of the peroxide (Hollaender, Greenstein and Jenrette, 1941). Butler and Conway (1953) studied the degrading action of hydrogen peroxide irradiated with ultraviolet light on DNA and simple model compounds. Evidence was obtained for the following modes of reaction: cleavage of the phosphate-sugar link, fission of the bond between sugar and base, and oxidation of the sugar and base moieties.

The depolymerizing action of hydrogen peroxide in the various systems has been attributed by Butler and coworkers to intermediate formation of the highly reactive hydroxyl and hydroperoxyl radicals.

If carcinogenesis involves template simplification, the hydroxyl radical may be only one of a number of reagents, including other radicals (Park, 1947 1950; Jensen, 1950; Butler, 1950, Greenstein, 1954d; Hirsch, 1956), capable of effecting this transformation. As an additional example, on the basis of a decrease in solution viscosity, nitrogen mustard appears to degrade nucleic acid (Butler et al., 1952; Greenstein, 1954d).

Role of Protein

It is not known with certainty whether or not protein is a crucial part of the template structure. If protein is intimately concerned, then similar considerations might be applied, in relation to simplification of the template, as were discussed in the case of nucleic acid. An attractive possibility would involve oxidative cleavage of disulfide crosslinks in the protein portion of the cell. Normally these crosslinks, which are part of the cystine structure, can be reversibly cleaved to cysteine residues. A reasonable oxidative route in the cancerous condition which may be of increased importance is the conversion to cysteinesulfinic acid and cysteic acid moieties. The significant point is the irreversible cleavage of crosslinks which are important in determining cell characteristics. A mass of evidence indicates that cancer protein has undergone changes of this or similar nature. Mason (1958) theorized that the associated protein exerts a protective action on nucleic acid, and that this effect is lost during carcinogenesis. In addition, interference by the carcinogen with normal association of nucleic acid with protein has been suggested (Haddow, 1957).

If protein does not constitute a vital part of the template, then the transformations discussed in the following sections can be considered a secondary consequence of cancer induction. In any case the experimental data provide additional support for the hydrogen peroxide hypothesis although it is recognized that other interpretations are possible.

Sulfur-containing constituents.—Many investigators have proposed that the sulfur-containing functional groups of cellular protein are concerned in some manner with the carcinogenic process. Shapiro and Eldjarn (1955a) previously suggested the irreversible oxidation of vital disulfide linkages as an important step in the mechanism of radiation damage (also see Eyring and Bowers, 1952) and, moreover, demonstrated (1955b) the conversion of cystamine to 2-aminoethanesulfinic acid and taurine by treatment with hydrogen peroxide. As a closer analogy, the oxidation in vitro with hydrogen peroxide of cystine to cysteic acid has been reported by Schöberl (1933).

Previously, statements concerning the amino acid composition of normal and

cancerous tissue have in most instances stressed their similarity. However, certain well-established differences in amino acid content may perhaps result from the high level of hydrogen peroxide which is deemed important in carcinogenesis, and which is reported (Rondoni and Cudkowiec, 1953) to exist in cancer cells. In a significant investigation, Greenstein and Leuthardt (1944) determined the cystine and cysteine content of extractable proteins derived from normal and neoplastic rat tissues. Although the analyses for total sulfur were essentially identical in the two cases, marked differences were noted in cystine, as well as cysteine, content. The values for the cancer transplants were as much as 40 to 50 percent below those for normal adult liver. In related work (Roberts et al., 1949), there were indications of a slight decrease in the level of cystine on conversion of normal skin to carcinoma.

A considerable amount of evidence from a number of laboratories points to a reduction in the sulfhydryl content of various protein constituents in the cancerous condition. From a study involving enzyme systems, Rondoni and Barbieri (1950) found that carcinogenic polynuclear compounds, such as benzpyrene and dibenzanthracene, inhibited the autolytic function of proteolytic enzymes, whereas noncarcinogenic hydrocarbons were without effect. The activity of these enzymes is intimately related to the presence of free sulfhydryl groups. Barron and co-workers (1949) have reported a number of interesting findings in this area of research. They demonstrated that this same type of enzyme, after inactivation by irradiation, could be reactivated to varying degrees by treatment with reducing agents. However, as the radiation dose was increased the destruction became increasingly irreversible. The latter result was interpreted by Shapiro and Eldjarn (1955a) on the basis of irreversible oxidation of the mercaptan groups beyond the disulfide state.

It is well established that a drop in serum sulfhydryl content occurs with the onset of neoplasia. For example, this effect has been observed for irradiation with x-rays and also for administration of nitrogen mustards (Shacter and Shimkin, 1950). Likewise, Wood and Kraynak (1953) noted a significant decline in the serum or plasma sulfhydryl content which persisted for a considerable time after intravenous injection of colloidal benzpyrene. Although it is known that cancer serum contains a subnormal amount of albumin, Schoenbach and his colleagues (1950) found that the decreased sulfhydryl levels still pertained even after correction for the reduction in albumin.

In an analysis for glutathione involving a comparison with normal rat liver, Greenstein (1942) reported a 24 percent lower content for the transplanted rat hepatoma, and Kinoshita (1938) a value 11 percent less in the case of the primary hepatoma. In another investigation, evidence was obtained (Rondoni and Boretti, 1947) for a decrease in the SH-content of water-soluble liver protein after intravenous injection of benzpyrene.

A number of the investigators cited have interpreted these results on the basis of destruction of sulfhydryl groups by oxidation, or, more commonly, by combination with the carcinogen.

Also pertinent to this discussion is the work of Ghosh and Lardy (1952) entailing alkali-treatment of acetone powders derived from various tissues. They reported that such treatment yielded elemental sulfur in considerably greater amount from tumorous as compared with normal tissue, and, on this basis, suggested a structure difference in the sulfur-containing proteins derived from the two sources. It is interesting that thiolsulfonic esters, products formed by partial oxidation of disulfides with hydrogen peroxide, are considered to yield sulfur as an intermediate product from their interaction with potassium sulfide (Connor, 1948).

Molecular weight. The occurrence of an irreversible oxidative cleavage of disulfide crosslinks would result in a decrease in the average molecular weight of the protein. Attack by hydrogen peroxide at other protein bonds also appears possible (Schumb, Satterfield and Wentworth, 1955c). It is well established that

the concentration in blood of proteinlike materials, which are not precipitated by heat or protein precipitants, increases in the blood of cancer patients (Winzler and Smyth, 1948). This condition is presumably caused by a rise in the level of low molecular weight proteins, proteoses or polypeptides (Greenstein, 1954e).

Excess hydrogen peroxide present in the body would also be expected to degrade other constituents, e.g., carbohydrate (Schumb, Satterfield and Wentworth, 1955d).

Coagulation.—The concept of a lower degree of crosslinking in cancer protein is consistent with the observation of Rondoni (1941), and also of Bassi and Bernelli-Zazzera (1954), that soluble cancer protein is more difficult to coagulate than corresponding normal protein. The same salt-soluble protein fraction was investigated for cancerous, normal and regenerating liver. There was a regular decrease in the ease of precipitation by heat with continued intake of *p*-dimethylaminoazobenzene, with a change in the opposite direction occurring in the case of regenerating liver. Furthermore, various investigators (Huggins, Cleveland and Jensen, 1950; Glass, Boyd and Dworecki, 1951) have reported that blood serum from a tumor-bearing host is more difficult to coagulate as compared with normal serum. In this connection, it is significant that Huggins and Jensen (1949) cite a definite correlation between the presence of sulfhydryl groups and turbidity formation.

Electrophoresis.—High concentrations of hydrogen peroxide would be expected to form acidic products, or products of increased polarity, not only from protein, but also from other body constituents, e.g., carboxylic acids from carbohydrates. There is general agreement among various investigators that soluble tumor protein possesses an electrophoretic pattern characterized by an increase in the proportion of faster-migrating components and a decrease in slower-moving ones, as compared with the corresponding normal protein (Sorof and Cohen, 1951; Eldredge and Luck, 1952; Hoffman and Schechtman, 1952; also see Ambrose, James and Lowick, 1956; Straumfjord and Hummel, 1959). This finding was interpreted by Sorof and Cohen (1951) as indicating a lower average isoelectric point for the soluble cancer protein. In addition, one of the components present in increased amounts was found to be a mucoprotein having an isoelectric point lower than pH 4 (Petermann and Hogness, 1948; Mehl, Golden and Winzler, 1949). It is very interesting that the acid protein gives a test reaction with toluidine blue similar to that characteristic of high molecular weight sulfuric acid esters (Petermann and Hogness, 1948).

Both an increase in polarity and a decrease in the molecular weight of cancer protein could account for the observed results in the electrophoresis experiments.

Related Considerations

Mutation.—Since many cancer-producing compounds are also mutagenic, it appears reasonable to classify carcinogenesis as an example of a mutation process in light of the concepts discussed. In many cases, both types of change may well proceed by a similar process: intermediary formation of hydrogen peroxide in high concentration. On this common basis, carcinogenesis would be regarded as producing a mutation characterized by a greater degree of dislocation.

Summary

It is believed that cancer may originate in the action of an agent which directly or indirectly generates hydrogen peroxide in abnormally high concentration. The subsequent stage is considered to be the formation of a template of simpler structure. Existing evidence is presented to support the proposed mode of action of hydrogen peroxide.

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THE SALAMANDERS OF SOUTHEASTERN OHIO*

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This is the first report resulting from a faunal survey of the vertebrates of southeastern Ohio, that has been conducted by the senior author for more than ten years. The junior author has concentrated on salamanders alone for the past three years and the accumulated information makes it advisable to set forth the results. This is especially appropriate for the salamanders, a group for which there is no modern list for the state of Ohio or any large segment thereof.

The literature on southeastern Ohio salamanders is not very extensive. The earlier works of Kirtland (1838), Smith (1882), Wilcox (1891), and Morse (1901, 1904), although purporting to include this section of the state, gave very few or no definite localities. In 1932, Walker and Weller reported *Gyrinophilus porphyriticus duryi* from Adams, Pike, and Highland counties; in 1933 Walker reported finding *Plethodon wehrlei* in Washington county and *Pseudotriton montanus* in Jackson county. Netting and Mittleman (1938) described *Plethodon richmondi* from Athens, Jackson, and Meigs counties. The Green Salamander (*Aneides aeneus*), although first reported for Ohio in 1941, was not found in our area until 11 years later (Gordon, 1952). Bishop (1941) described *Pseudotriton montanus diastictus* as a new subspecies occurring in southeastern Ohio, although Mittleman and Gier (1948) maintained that an isolated population of *P. m. montanus* occurred in Jackson, Gallia, and Athens counties. In 1942, Mittleman described *Gyrinophilus porphyriticus inagnoscus* from southeastern Ohio and, in 1949, *Eurycea bislineata rivicola* as the race of the Two-lined Salamander occurring west of the Alleghany Mountains.

The area included within our definition of southeastern Ohio comprises Washington, Morgan, Athens, Hocking, Ross, Pike, Scioto, Lawrence, Gallia, Meigs, Jackson, and Vinton counties. This area essentially includes all of the unglaciated portion of the Southern Allegheny Plateau. In the following list, each locality record is ascribed to its county and following the locality, in parentheses, is an abbreviation denoting the disposition of the specimen or whence the information concerning the record was obtained. The abbreviations used are: (OUVC) Ohio University Vertebrate Collection; (OSM) The Ohio State Museum; (MC) Marietta College; (CM) Carnegie Museum; (CMNH) Cleveland Museum of Natural History; (OWU) Ohio Wesleyan University; (UIMNH) University of Illinois Museum of Natural History; (NBG) N. Bayard Green; (GMD) George McDuffie; (JC) John M. Condit; (DMD) D. M. Dennis; (RB) Ronald Brandon. All specimens were examined personally unless otherwise noted. References from the literature are cited in the usual manner. Our nomenclature is from the 6th edition of the Check List of North American Amphibians and Reptiles and we use the common names prepared by the Committee on Herpetological Common Names (1956).

The provenance of each specimen is indicated on the outline maps as accurately as possible. Quotation marks around the symbol indicate that no specific locality within that county has been recorded.

Considerable effort has gone into the accuracy and completeness of these distributional data. It becomes apparent that even within a small area, such as

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these dozen counties, salamanders are not uniformly dispersed. Even though intensive collecting has not been possible in all areas, there is no doubt that many townships will remain blank for lack of suitable habitat. It is hoped that these detailed data will be of value years from now for comparative purposes; it would have been exciting to us had it been possible to compare the present day distribution of these salamanders with that of 80 years ago.

A total of 19 species of salamanders has been found within this area and this total is very likely complete. The details of distribution may alter to some degree, principally by the addition of new records that will fill the gaps existing among already recorded collecting sites, but it is now evident that certain species (*A. aeneus*, *P. wehrlei*, for example) have, and will continue to have, very restricted distribution and others (*E. bislineata*, *D. fuscus*) are widespread. Undoubtedly some (*C. alleganiensis*) have had their distribution curtailed.

A list such as this should indicate the relative abundance of the different species, but no method has been devised to evaluate this statistic for salamanders. Some species are easier to find than others but whether this reflects a true picture of their abundance is doubtful. Nevertheless, based on distributional records, frequency in collections, and personal observations, the species may be grouped as follows:

Very Common	Fairly Common	Uncommon	Rare
<i>D. viridescens</i>	<i>N. maculosus</i>	<i>C. alleganiensis</i>	<i>A. texanum</i>
<i>P. cinereus</i>	<i>A. opacum</i>	<i>H. scutatum</i>	<i>A. aeneus</i>
<i>P. glutinosus</i>	<i>A. jeffersonianum</i>	<i>P. ruber</i>	<i>P. wehrlei</i>
<i>D. fuscus</i>	<i>A. maculatum</i>	<i>P. montanus</i>	
<i>E. bislineata</i>	<i>P. richmondi</i>	<i>G. porphyriticus</i>	
	<i>E. longicauda</i>		

In the first category are those species which can be found in many places most of the time; the second group includes those which are restricted to a greater degree in habitat or seasonal occurrence but which, at the right place and at the right time, may be found in reasonable numbers; the third group are those which are never found in any great numbers; the last group includes the three species whose distribution is narrowly limited.

Miscellaneous data on breeding and other aspects of life history have been included whenever the information was available or deemed worthy of recording. Additional details on ecology, taxonomy, and synonymy can be found in Brandon's (1958) report.

Cryptobranchus alleganiensis alleganiensis Daudin, Hellbender.—ATHENS Co. Hocking R., Athens (OU Zool. Dept.). SCIOTO Co. Scioto Brush Crk., near Henley (OSM 220). WASHINGTON Co. Muskingum R., Marietta (MC) (fig. 1).

Necturus maculosus maculosus Rafinesque, Mudpuppy.—ATHENS Co. Hocking R., Athens (OUVC 1568, 2764, 2765, 2771, 2854); near Stewart (OUVC 2896); mouth of Hocking R., (fide W. Mansfield). GALLIA Co. Raccoon Crk. (OSM 885). HOCKING Co. Hocking R., Logan (RB 88). MEIGS Co. Middle Shade R., Orange Twp. (OUVC 3998); Leading Crk., Salisbury Twp. (RB). WASHINGTON Co. Muskingum R., Marietta (MC) (fig. 2).

Ambystoma jeffersonianum Green, Jefferson Salamander.—ATHENS Co. 2½ mile e of Athens (OUVC 1793); between Chauncey and Nelsonville on US 33 (OUVC 2929); near Stewart (OUVC 2894); Guysville (OUVC 3003); East Branch Sunday Crk., Gloucester (OSM 664); Waterloo Wildlife Area (RB 55); Stroud Run, Sec. 13, Canaan Twp. (RB 58). HOCKING Co. Silica Hollow, Goodhope Twp. (OSM 1058); East Buckrun, Goodhope Twp. (OUVC 3956); Benton Twp. (OSM 391); ne Sec. 11, Falls Twp. (OUVC 3323-24). JACKSON Co. no particular locality (OUVC 107) (OSM 985). MORGAN Co. ½ mile se of Bishopville (OUVC 1794). PIKE Co. Coopersville (OSM 884). ROSS Co. 2 mile w of Chillicothe, Scioto Twp. (OUVC

3707). Scioto Co. near Bear Lake, Shawnee Forest (GMD). VINTON Co. Wolf Run, se Sec. 34, Elk Twp. (OUVC 3332); near Lake Hope (RB 94) (fig. 3).

The earliest date when eggs were found was on February 9, 1957, when several clusters were observed in a hilltop pond in a deciduous woods near Logan, Hocking County. This pond was approximately five feet square and about two feet deep near the center. Egg masses were attached to petioles of oak leaves, stems of various herbaceous plants, and small tree stems that had fallen into the pond. The number of eggs in several masses varied from 8 to 55, with an average number of 22. On March 2, 1957, 22 adult Jefferson Salamanders, 17



FIGURE 1. The distribution of *Cryptobranchus alleganiensis* (open circles); *Ambystoma opacum* (solid dots); *Hemidactylium scutatum* (crosses).



FIGURE 2. The distribution of *Necturus maculosus* (crosses); *Plethodon glutinosus* (solid dots).

males and five females, were taken from the above pond. The males ranged from 150 to 200 mm in length; the females, 160 to 180 mm. Three clusters of eggs were found on February 23, 1957, in a small roadside ditch near Canaanville, Athens Co. Other eggs were collected on March 9, 1957, in a roadside swampy area near Waterloo Wildlife Area, Athens Co., and on

March 17, 1957, a cluster was found in a small, spring-fed pool on an open pastured hillside near Canaanville.

Ambystoma maculatum Shaw, Spotted Salamander.—ATHENS Co. Second St., Athens (OUVC 1805); Carbondale (OUVC 1556); Waterloo Wildlife Area (RB 49, 55). GALLIA Co. near Rodney, Springfield Twp. (OSM 360). HOCKING Co. near Logan, Falls Twp. (RB 13), East Buckrun, Goodhope Twp. (CMNH ZF1507). JACKSON Co. no particular locality (OUVC



FIGURE 3. The distribution of *Ambystoma jeffersonianum* (crosses); *Diemictylus viridescens* (solid dots).



FIGURE 4. The distribution of *Eurycea longicauda* (crosses); *Ambystoma maculatum* (solid dots).

105); Jackson Twp. (OUVC 475); White's Gulch, Liberty Twp. (OUVC 1543). PIKE Co. cave near Byington (OSM 554). ROSS Co. 2 mile w of Chillicothe, Scioto Twp. (OUVC 3706). SCIOTO Co. Shawnee (GMD). VINN Co. Bolster Hollow, near Lake Hope (RB 61a); King Hollow, n of Lake Hope (RB 96); Ohio 356 near US 50 (RB 49) (fig. 4).

Egg masses were collected on March 6, 1957, March 22, 1957 and April 11, 1958, all from

near Lake Hope, Vinton County. The clusters were attached to the stems of standing herbaceous plants from a few inches to almost a foot below the water surface. The number of eggs in masses varied from 60 to 90.

Ambystoma opacum Gravenhorst, Marbled Salamander.—ATHENS Co. Athens (OUVC 802, 2909); near Armitage (RB 56); Waterloo State Forest (OUVC 2402, 2947); Stroud Run, Canaan Twp. (RB a64); Sec. 7, Lee Twp. (OUVC 2812); near Canaanville (RB 48). GALLIA Co. Sec. 13, Huntington Twp. (RB 57a). HOCKING Co. East Buckrun, Goodhope Twp. (OUVC 3955); Ash Cave (OUVC 3000); near Coonville (RB 75). JACKSON Co. Jackson Twp. (OUVC 476); Rock Run (OSM 921); Sec. 17, Liberty Twp. (OUVC 3699). LAWRENCE Co. no particular locality (NBG). Ross Co. 2 mile w of Chillicothe, Scioto Twp. (OUVC 3677). SCIOTO Co. Shawnee Forest (GMD); Portsmouth (Morse, 1901). VINTON Co. Lake Hope (OUVC 2991); near Zaleski (RB 96); Vales Mills (RB 103).

Although adults were seldom collected except in the autumn, the larvae remained in ponds throughout the winter and into the next spring, and were easily found and collected during this time. In one breeding pond near Waterloo Wildlife Area, Athens County, hundreds of larvae were seen swimming about all winter (fig. 1).

Ambystoma texanum Matthes, Small-mouthed Salamander.—HOCKING Co. Clear Crk. (OSM 907.2). WASHINGTON Co. 6 mile w of Marietta (OUVC 1190); Squaw Hollow, Marietta (OUVC 502).

To our knowledge, the Small-mouthed Salamander has been found in only two areas in southeastern Ohio, although it appears to be more common to our north in Fairfield County and to our west in the Cincinnati area (fig. 5).



FIGURE 5. The distribution of *Ambystoma texanum* (crosses); *Plethodon richmondi* (solid dots).

Diemictylus viridescens viridescens Rafinesque, Red-spotted Newt.—ATHENS Co. Sell's Park, Athens (RB 23); 2 mile e of Athens (OUVC 63); 3 mile ne of Athens (OUVC 1570); Waterloo Wildlife Area (RB 53); Carbondale Forest (RB 54). GALLIA Co. Vinton, Sec 24, Huntington Twp. (OUVC 3339); no particular locality (OSM 922). HOCKING Co. Salt Crk., 1 mile w of Ash Cave (OUVC 1766); Cantwell Cliffs (OSM 678); Laurelville-Enterprise Rd (OSM 571); Conkle's Hollow (OSM 702); Neotoma, Goodhope Twp. (OSM 295); East Buckrun, Goodhope Twp. (CMNH ZF1506). JACKSON Co. Lake Alma (OUVC 1806); Lake Jackson (OUVC 2498); no particular locality (OUVC 472, 473); Rock Run (OSM 921). LAWRENCE Co. Olive Furnace (OSM 839); Dean Forest, Decatur Twp. (OSM 141). MEigs Co. Sec. 18, Rutland Twp. (OUVC 218, 222-24); Salem Twp. (OSM 141). MORGAN Co. York Twp. (OSM 864). PIKE Co. Waverly

(OSM 88); near Jasper (OSM 58). Ross Co. Tar Hollow, Colerain Twp. (OUVC 3359); Bainbridge (OSM 1401); Spruce Hill, Twin Twp. (OSM 315). Scioto Co. Lucasville (OSM 199); Shawnee Forest (GMD); nw Rush Twp. (OUVC 3889). Vinton Co. King Hollow, Brown Twp. (RB 49); Lake Hope (OUVC 2397, 2398); near Eagle Mills (OSM a486); Vales Mills (RB 103). WASHINGTON Co. Fairfield Twp. (OSM 619); Belpre Twp. (OSM 586); Squaw Hollow, Marietta (MC) (fig. 3).

On two occasions spring migrations have been observed near Lake Hope in Vinton County. On the night of March 19, 1954, during a warm rain, dozens of newts were observed crossing Ohio Route 278 between Lake Hope and Bolster Hollow. Many crushed individuals were found on the road. On the night of March 29, 1958, several newts were observed crossing the same highway near Zaleski. On May 4, 1958, several pairs of adults were seen clasping in a roadside ditch in Vales Mills, Vinton County. Since the females were quite plump, evidently swollen with eggs, two pairs were brought back to the laboratory. The next day, May 5, the two females laid some eggs, attaching them to blades of grass. These eggs hatched on May 15 and 16. On August 3, 1958, several fully grown larval newts were collected from a roadside ditch near Waterloo Wildlife Area. Within three days after being brought back to the laboratory, all the larvae transformed.

Desmognathus fuscus fuscus Rafinesque, Northern Dusky Salamander.—ATHENS Co. 1 mile se of Athens (OUVC 81, 88, 89); 1½ mile se of Athens (OUVC 432, 433, 801); 1 mile e of Athens (OUVC 1763, 2223); 3 mile ne of Athens (OUVC 1303); 2½ mile ne of Athens (OUVC 1779); 1½ mile e of Athens (OUVC 97, 100); Athens (OUVC 108, 1763); Rock Rille, Athens (OUVC 1554); Sec. 31, Lodi Twp. (OUVC 2431); Sec. 30, Lodi Twp. (OUVC 423, 424, 1399); ne Sec. 18, Lodi Twp. (RB 21); Lodi Twp. (OUVC 1118); Sec. 8, Lodi Twp. (OUVC 83-86); Shade, Lodi Twp. (OUVC 1127-30); Beaumont, Dover Twp. (OUVC 2319); Waterloo Twp. (OUVC 1877); Waterloo Forest (OUVC 2208); 1 mile s of Waterloo (OUVC 1135); Carbondale (OUVC 90-92, 500, 501, 1148, 1149, 2203); ½ mile n of The Plains (OUVC 82, 87); Trimble Twp. (OUVC 1772); se of Nelsonville (OUVC 1769); Troy Twp. (OSM 653); 1 mile sw of Athens (OUVC 1131); Stroud Run, nw Canaan Twp. (RB 37, 74); Hidden Valley, Sec. 13, Canaan Twp. (RB 58); sw Sec. 25, Rome Twp. (RB 10); Coolville (RB 20); ne Troy Twp. (RB 67). GALLIA Co. nc Sec. 14, Huntington Twp. (OUVC 3343); Springfield Twp. (OSM 360). HOCKING Co. Ash Cave (OUVC 1886) (OSM 273); near Rock House (OSM A662); Cantwell Cliffs (OSM 636); Conkle's Hollow (OSM 702); Neotoma (OSM 165); Salt Crk. Benton Twp. (OUVC 1550) (OSM A645); Sec. 33-34, Benton Twp., 3 mile se of Bloomingville (OUVC 1873); Sec. 19, Goodhope Twp., 3 mile e of Revenge (OUVC 1975); Goodhope Twp. (OSM 89); Salt Creek Twp., near Haynes (OSM 148); Sec. 9, Laurel Twp. (OSM 154); Logan (RB 32); n of Logan, Marion Twp. (RB a32); Old Man's Cave (RB 82); near Blackjack, Big Pine Rd. nw Washington Twp. (RB 43). JACKSON Co. Jackson Twp. (OUVC 523-26, 474, 483, 384); Jackson (OUVC 1188); no particular locality (OUVC 1541) (OSM 833); White's Gulch, Liberty Twp. (OSM 673); Sec. 20, Jefferson Twp. (OUVC 3879). LAWRENCE Co. Decatur Twp. (OSM 358); Dean Forest, Decatur Twp. (OSM 141); Sec. 14, Washington Twp. (OUVC 4002). MEIGS Co. Darwin, Sec. 16, Bedford Twp. (OUVC 93-96, 2230); Pomeroy (OSM 433); Long Bottom (OUVC 3225); Salem Twp. (OSM 359); 2 mile e of Bashan (OSM 818); Sec. 16, Scipio Twp. (OUVC 3866); Forked Run Lake, Olive Twp. (RB 83). MORGAN Co. York Twp. (OSM 864); 6 mile e of McConnellsville (OUVC 2228); se Sec. 17, Malta Twp. (OUVC 3353); Malta Twp. (OSM 398); Kill Deer, Deerfield Twp. (OSM 397). PIKE Co. near Byington, Millin Twp. (OSM 339, 554); "Big Cave," Millin Twp. (OSM 82); se Perry Twp. (OUVC 3443). ROSS Co. near Tucson, Harrison Twp. (OSM A487); Chillicothe (OUVC 2979) (Morse, 1904); Tar Hollow (RB 87); ne Paint Twp. (OUVC 3856). SCIOTO Co. Roosevelt Lake (OUVC 3093-4, 3097-9); Shawnee Forest, Union Twp. (OSM 432); sw Nile Twp. (OUVC 3885); Sec. 30, Madison Twp. (OUVC 3892). VINTON Co. Lake Hope (OUVC 1893); Brown Twp. (RB 85); Knox Twp. (OSM 280); Sec. 4, Richland Twp. (OUVC 3774); Sec. 4, Jackson Twp. (OUVC 3818); s Sec. 7, Eagle Twp. (OUVC 3822). WASHINGTON Co. Marietta Twp. (OUVC 1139-41); Squaw Hollow, Sec. 35, Marietta Twp. (OUVC 2463) (MC); off Indian Run (MC); Belpre Twp. (OSM 586); Lawrence Twp.

(OSM 321); e Sec. 11, Decatur Twp. (RB 69b); near Cutler (RB a44); Sec. 16, Ludlow Twp. (OUVC 3686) (fig. 6).

On January 10, 1957, a hibernating group of fifty adult, several juvenile, and ten larval duskies was discovered in a swampy alder woods near Beaumont, Athens County. After a large, rotten tree stump had been torn out of the damp soil, the salamanders were exposed in depressions in the stump and in soil pockets under and around the stump.



FIGURE 6. The distribution of *Desmognathus fuscus* (solid dots); *Aneides aeneus* (cross).

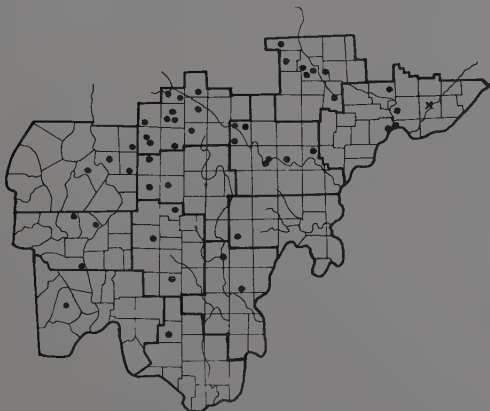


FIGURE 7. The distribution of *Plethodon cinereus* (solid dots); *Plethodon wehrlei* (cross).

Plethodon cinereus cinereus Green, Red-backed Salamander.—ATHENS Co. Athens (OUVC 2227); Sell's Park, Athens (RB); Stroud Run, Canaan Twp. (RB 66d, 74); Rome Twp. (OUVC 3348); Buchtel (OUVC 2206); Enderline Forest, near Carbondale (RB 101); Nelsonville (Morse, 1904). GALLIA Co. sw Sec. 24, Huntington Twp. (OUVC 3858); near Rodney, Springfield Twp. (OSM 360). HOCKING Co. Ash Cave (OUVC 2321); near Rock House (OSM A662); Sec. 35, Marion Twp. (OUVC 3199, 3200); Salt Creek (OSM A645); Salt Creek Twp. (OSM 148); near

Haynes (OSM 214); Clear Creek, Goodhope Twp. (OSM 89); Laurel Twp. (OSM 154); Laurelville (OSM 152); Falls Twp. (RB a34); Conkle's Hollow (RB 43); near Blackjack, Big Pine Rd., Washington Twp. (RB 43); se Sec. 24, Goodhope Twp. (RB 7). JACKSON Co. Lake Jackson, Jefferson Twp. (OUVC 2497); White's Gulch, Liberty Twp. (OUVC 2297) (OSM 673); Washington Twp. (OSM 408). LAWRENCE Co. Decatur Twp. (OSM 358). MEIGS Co. Salem Twp. (OSM 359). MORGAN Co. Malta Twp. (OSM 398); se Sec. 17, Malta Twp. (OUVC 3349-50); Malta (OSM 498); Kill Deer, Deerfield Twp. (OSM 397); York Twp. (OSM 864); along Muskingum, Meigsville Twp. (OSM 588); Roxbury (OSM 866); no particular locality (Morse, 1904). PIKE Co. near Waverly (OSM 59); near Ross Co. line on US 23, Pee Pee Twp. (OSM 180); Starr Hollow, near Coopersville (OSM 587). ROSS Co. Sec. 26, Harrison Twp. (OUVC 3857); Mt. Eyes, Springfield Twp. (OSM 344); Tar Hollow, Colerain Twp. (OUVC 3360); Chillicothe (Morse, 1904). SCIOTO Co. Union Twp. (OSM 200). VINTON Co. se Sec. 8, Richland Twp. (OUVC 3820); Harrison Twp. (OSM 822); near Salt Creek (OSM 472). WASHINGTON Co. Lowell (OSM 756); Marietta (OSM 620); ne Sec. 3, Warren Twp. (OUVC 3286); Sec. 35, Marietta Twp. (OUVC 2464); e Muskingum Twp. (OUVC 3355) (fig. 7).

The Red-backed Salamander is the commonest terrestrial salamander in most areas of southeastern Ohio. However, in some parts of Athens County, the Ravine Salamander, *P. richmondi*, is abundant to the exclusion of *P. cinereus*. The latter is abundant in Sell's Park, Athens, but *richmondi* has never been collected there. Conversely, in the Athens State Hospital woods, 2 miles southwest of Sell's Park, *richmondi* is very common whereas *cinereus* can not be found.

Plethodon glutinosus glutinosus Green, Slimy Salamander.—ATHENS Co. Athens (OUVC 1764, 2461); Rock Riffle, 1½ mile se of Athens (OUVC 65, 67); 2½ mile ne of Athens (OUVC 1778); Carbondale (OUVC 1146, 1147); Waterloo Forest (OUVC 71, 72, 1197); Buchtel (OUVC 2205); 2 mile n of Buchtel (OUVC 1393, 1395); Buffalo Beat, 1½ mile se of Buchtel (OUVC 1761); Stroud Run, Canaan Twp. (RB 84); 7 mile se of Athens, Lodi Twp. (OUVC 1403). GALLIA Co. sw Sec. 24, Huntington Twp. (OUVC 3859); Cadmus, Walnut Twp. (OUVC 3860); 1 mile s of Vinton (RB 86); no particular locality (OSM 833). HOCKING Co. Ash Cave (OUVC 3301-02); near Rock House (OSM A622); Rock House (RB 5); Salt Creek (OSM A645); Salt Creek Twp. (OSM 214); near Haynes, Salt Creek Twp. (OSM 148); Clear Creek (OSM 195); Goodhope Twp. (RB a4); Old Man's Cave (RB 14); Sec. 9, Laurel Twp. (OSM 154); sw Sec. 35, Marion Twp. (RB 15). JACKSON Co. Sec. 14, Liberty Twp. (OUVC 2992, 2993); White's Gulch, Liberty Twp. (OSM 673); Jackson Twp. (OUVC 489); no particular locality (OUVC 471). LAWRENCE Co. Hecla (OSM 838); near Crown City, 3 mile from Gallia Co. line (CM). MEIGS Co. Bedford Twp. (OUVC 74); Darwin (OUVC 2231); 2 mile s of Bashan (OSM 818); no particular locality (OSM 964). PIKE Co. 1 mile s of Piketon (OUVC 2408); Starr Hollow (OSM 587); near Waverly (OSM 59). ROSS Co. Copperas Mt., Twin Twp. (OSM 257); Spruce Hill, Twin Twp. (OSM 315); Sugar Loaf Mt., Green Twp. (OSM 345). SCIOTO Co. Union Twp. (OSM 200). VINTON Co. Knox Twp. (OSM 280); Waterloo Forest (OUVC 1197); Bolster Hollow, Brown Twp. (CMNH ZF1494). WASHINGTON Co. ne Sec. 3, Warren Twp. (OUVC 3285); Squaw Hollow, Sec. 35, Marietta Twp. (OUVC 3288) (MC); along Little Muskingum R., Lawrence Twp. (OSM 830); Lawrence Twp. (OSM 321); Lowell (OSM 756); Sec. 16, Ludlow Twp. (OUVC 3687); nw Sec. 13, Grandview Twp. (RB 102) (fig. 2).

Plethodon richmondi Netting and Mittleman, Ravine Salamander.—ATHENS Co. Athens (OUVC 479-82, 2226); ½ mile e of Athens (OUVC 1408); 1 mile se of Athens (OUVC 30, 434, 435, 437, 438, 1551); 1 mile e of Athens (OUVC 1765); 1 mile sw of Athens (OUVC 25, 1137); ½ mile sw of The Plains (OUVC 1559); Alexander Twp. (OUVC 26, 1123, 1124); Lodi Twp. (OUVC 1401); Sec. 30, Lodi Twp. (OUVC 425-29); Sec. 13, Canaan Twp. (OUVC 3345). JACKSON Co. White's Gulch (OUVC 1540); Jackson Twp. (OUVC 485-87); no particular locality (OSM 966). LAWRENCE Co. Sec. 30, Windsor Twp. (OUVC 3863); Decatur Twp. (OSM 358); near Crown City (CM 14102 04). MEIGS Co. Sec. 16, Bedford Twp. (OUVC 27-29); 1 mile s of Pratt's Fork (OUVC 2216); Darwin (OUVC 2232); Forked Run State Park, Olive Twp. (OUVC 3566). PIKE Co. near Waverly (OSM 59); near Ross Co. line, US 23, Pee Pee Twp. (OSM 180); Benton Twp. (OUVC 3442). ROSS Co. 2 mile w of Chillicothe, Scioto Twp. (OUVC

3676). SCIOTO Co. Shawnee Forest (GMD). WASHINGTON Co. Squaw Hollow, Sec. 35, Marietta Twp. (OUVC 2466) (MC); 8 mile nw of Marietta (OUVC 1395); nw Sec. 13, Grandview Twp. (OUVC 3690); Sec. 5, Lawrence Twp. (OUVC 3688-89) (fig. 5).

Three adult female Ravine Salamanders collected from the Athens State Hospital grounds on March 26, 1958, were opened and examined. One specimen, 12.5 cm from snout to tail tip, contained eight well-developed eggs, five in one uterus, three in the other. The eggs measured 2.3 mm to 3.2 mm in diameter. In the stomach were five sowbugs and some semidigested material. The second, 12.4 cm long, contained eggs measuring from 2.3 mm to 3.7 mm in diameter, five in one uterus, four in the other. The stomach contained one spider, one rove beetle, one unidentified insect, and seventeen sowbugs. The third, 8.4 cm long, had no developed eggs, but the stomach contained one rove beetle and nineteen sowbugs.

Plethodon wehrlei Fowler and Dunn, Wehrle's Salamander.—WASHINGTON Co. along Little Muskingum R., Lawrence Twp. (OSM 830) (fig. 7).

Wehrle's Salamander has been collected from only two localities in Ohio: along Sunfish Creek in Monroe County and along the Little Muskingum in Washington County (Walker, 1933).

Hemidactylium scutatum Schlegel, Four-toed Salamander.—GALLIA Co. no particular locality (OSM 883). HOCKING Co. Old Man's Cave (OUVC 1101, 1102) (OSM 799). JACKSON Co. White's Gulch, Liberty Twp. (OSM 673); no particular locality (OSM 833). SCIOTO Co. Shawnee Forest (GMD) (fig. 1).

Gyrinophilus porphyriticus duryi Weller, Kentucky Spring Salamander.—PIKE Co. Coopersville (CM 5850, 5851); 2 mile n of Coopersville (OSM 676); Richardson Hollow, nw Benton Twp. (OUVC 3438); Kincaid Crk., se Perry Twp. (OUVC 3439); Pike State Park (OUVC 3573); 1 mile n of Morgantown (OUVC 3946). SCIOTO Co. Roosevelt Lake, Shawnee S.F. (OUVC 3085); Roosevelt Preserve, Nile Twp. (OSM 963). Weller (1930), Walker (1932), and Mittleman (1942) record specimens from Pike and Scioto Counties (fig. 8).



FIGURE 8. The distribution of *Gyrinophilus porphyriticus porphyriticus* (solid dots); *inagnoscus* (half-dots); *duryi* (crosses).

Gyrinophilus porphyriticus inagnoscus Mittleman, Ohio Spring Salamander.—ATHENS Co. Carbondale Forest (UIMNH 36177). HOCKING Co. Salt Creek, 4 mile sw of Bloomingville, Goodhope Twp. (Mittleman, 1942, but cf. Condit, 1958); Clear Creek Canyon, Goodhope Twp. (OWU 3264); Conkle's Hollow State Park (OUVC 3175); Cantwell Cliffs State Park (OUVC 3705, 3896); (OWU 3265); Sec. 27, Laurel Twp. (OUVC 3871); Sec. 32, Marion Twp. (RB 33).

Although the latter is a larval specimen, it is considered to be *inagnoscus* because of its nearness to the Hocking parks (fig. 8).

Gyrinophilus porphyriticus porphyriticus Green, Northern Spring Salamander.—ATHENS Co. Stewart (OUVC 2966). PIKE Co. Pike State Park (OUVC 3256). WASHINGTON Co. ne Sec. 3, Warren Twp. (OUVC 3287, 3294); Sec. 35, Marietta Twp. (OUVC 3292); Squaw Hollow (UIMNH 36169-75) (fig. 8).

The status of *Gyrinophilus* in southeastern Ohio is, to our minds, still unsettled. Brandon (1958), on the basis of specimens available to him, felt that a) *G. p. duryi* is a well defined race occurring in the extreme western portion of our area, with no evidence of intergradation to the east; b) *G. p. inagnoscus* is a poorly defined race, differing from eastern representatives only in pigmentation, and restricted in distribution to Hocking and western Athens Counties; c) the rest of southeastern *Gyrinophilus* are closer to *porphyriticus* than to *inagnoscus* even though Mittleman (1942) considers specimens from Washington County to be *inagnoscus*. Paratypes of *inagnoscus* (UIMNH 36173-75) are considered by us to be closer to *porphyriticus*.

One specimen (OUVC 3256) from Pike County, brought to us and substantiated by William Price, Park Naturalist of the Ohio Division of Parks, is definitely not *duryi* but exactly like the eastern forms. This specimen had been regurgitated by a common garter snake (*Thamnophis sirtalis*). From its excellent condition it could not have been long in the snake's digestive tract and we can only surmise that the salamander had been captured by the snake in Pike State Park. If this assumption is correct, the status of *duryi* as a subspecies of *porphyriticus* needs to be re-examined.

Pseudotriton montanus diastictus Bishop, Midland Mud Salamander.—ATHENS Co. Sec. 20, York Twp. (OUVC 3742). GALLIA Co. no particular locality (OSM 883.5, 919.1, 919.2). JACKSON Co. Washington Twp. (OSM A401, 406); Liberty Twp. (OSM 294.18, A981, 673). LAWRENCE Co. no particular locality (NBG). ROSS Co. Sec. 12, Harrison Twp. (OUVC 3816). SCIOTO Co. Roosevelt State Park (OUVC 2932); Shawnee Forest (GMD). VINTON Co. Lake Hope (OUVC 2969, 3265); King Hollow, near Lake Hope (RB 61b) (fig. 9).

Mittleman and Gier (1948) are of the opinion that some of the *Pseudotriton montanus* in southeastern Ohio are not *diastictus* but an isolated population of *P. m. montanus*, the Eastern Mud Salamander, which occurs to the south and east of Ohio. According to these writers, specimens from Jackson, Gallia, and Athens counties are *P. m. montanus*, those from Jackson and Adams counties are *P. m. diastictus*, and some from Jackson County, a specimen from Ross County, and a specimen from Scioto County, are considered to be intergrades.

However, since specimens in our collection from Athens, Vinton, and Scioto counties are, in our opinion, definitely *P. m. diastictus*, and since the specimens in The Ohio State Museum that were examined by Mittleman and Gier appear to us to be the same, all southeastern Ohio specimens are considered to be *diastictus*.

Pseudotriton ruber ruber Sonnini, Northern Red Salamander.—ATHENS Co. Athens (OUVC 2900); 2 mile ne of Athens (OUVC 2144); Stroud Run, Sec. 23, Canaan Twp. (OUVC 3223, 3279); Sec. 33, Canaan Twp. (OUVC 3935); Beaumont (OUVC 3177); se Sec. 10, Dover Twp. (OUVC 2544). GALLIA Co. near Rodney, Springfield Twp. (OSM 360). HOCKING Co. Neotoma (OSM 239); Clear Creek (OSM 54, 293); Cantwell Cliffs (OSM 678); Sec. 26, Goodhope Twp. (JC). JACKSON Co. White's Gulch, Liberty Twp. (OSM 673). SCIOTO Co. Roosevelt Game Preserve, Nile Twp. (OSM 201, 735); Shawnee Forest (GMD). WASHINGTON Co. Belpre Twp. (OSM 586) (fig. 9).

Aneides aeneus Cope and Packard, Green Salamander.—LAWRENCE Co. Burlington (NBG) (Gordon, 1952) (OUVC 4006-8) (fig. 6).

Eurycea bistlineata rivicola Mittleman, Midwest Two-lined Salamander.—ATHENS Co. Athens (OUVC 2426) (RB 151); 5 mile e of Athens (RB 89); Stroud Run, Canaan Twp. (RB 6); Sec. 13, Canaan Twp. (OUVC 3344); near Carbondale (RB 106); Beaumont, Dover Twp. (OUVC 2318); Troy Twp. (OSM 653); Fourmile Run, n Sec. 32, Troy Twp. (OUVC 3289-91); Sec. 31, Lodi Twp. (OUVC 2432); sw of Shade (RB 79); 3 mile n of Coolville (RB 20). GALLIA Co. nw Sec. 25, Greenfield Twp. (OUVC 3865); near Rodney, Springfield Twp. (OSM 360). HOCKING

Co. Ash Cave (OUVC 2320); Goodhope Twp. (OSM 89); sw Sec. 18, Goodhope Twp. (RB 68); Sec. 25, Goodhope Twp. (RB 2); Clear Creek (OSM 239, 197); Sec. 9, Laurel Twp. (OSM 154); Rock House (OSM A662); Cedar Falls (RB a31); Conkle's Hollow (RB 78); Sec. 27, Marion Twp. (OUVC 3201); near Haynes, Salt Creek Twp. (OSM 148); Salt Creek (OSM A645); near Blackjack, Big Pine Rd., Washington Twp. (RB 43); Cantwell Cliffs State Park (RB 17); Lake Hocking (RB 26); Logan (RB 32). JACKSON Co. Sec. 14, Liberty Twp. (OUVC 2995-96); White's Gulch, Liberty Twp. (OSM 294); Rock Run (OSM 833); Sec. 11, Hamilton Twp. (OUVC 3880). LAWRENCE Co. Sec. 14; Windsor Twp. (OUVC 3861); Sec. 18, Windsor Twp. (OUVC 3862); Manker, Sec. 2, Lawrence Twp. (OUVC 3864); Decatur Twp. (OSM 358); Burlington (OUVC 4005). MEIGS Co. 1 mile s of Pratts Fork (OUVC 2215); Salem Twp. (OSM 359);



FIGURE 9. The distribution of *Pseudotriton ruber* (solid dots); *Pseudotriton montanus* (crosses).

Maloon's Run, nw Sec. 7, Salem Twp. (OUVC 3340-42); 2 mile e of Bashan (OSM 818); Sec. 35, Rutland Twp. (OUVC 3868); Sec. 12, Sutton Twp. (OUVC 3869); Long Bottom (OUVC 3226). MORGAN Co. York Twp. (OSM 864); se Sec. 17, Malta Twp. (OUVC 3351-52); Malta Twp. (OSM 398); Kill Deer, Deerfield Twp. (OSM 397). PIKE Co. Starr Hollow, w of Coopersville (OSM 597); n of Waverly (OSM 59); near Ross Co. line and US 23, Pee Pee Twp. (OSM 180); Pebble Twp. (OSM 521); nw Benton Twp. (OUVC 3441); Sec. 31, Beaver Twp. (OUVC 3881). ROSS Co. near Tucson, Harrison Twp. (OSM A487); Jintown Hollow, Paxton Twp. (OUVC 3440); 1 mile sw of Austin (OUVC 3853); Porter Hollow Rd., s Concord Twp. (OUVC 3852); 3 mile w of Bourneville, Twin Twp. (OUVC 3854); ne Paint Twp. (OUVC 3853). SCIOTO Co. Roosevelt Lake, Shawnee S.F. (OUVC 3100) (OSM 558); sw Nile Twp. (OUVC 3884); w Brush Creek Twp. (OUVC 3887); Harrison Twp. (OSM 384); 5 mile n of Wheelersburg (OSM 449); Sec. 30, Madison Twp. (OUVC 3891). VINTON Co. Sandy Run, Sec. 10, Brown Twp. (OUVC 2522-24); Knox Twp. (OSM 280); Sec. 4, Jackson Twp. (OUVC 3817); se Sec. 8, Richland Twp. (OUVC 3819); s Sec. 7, Eagle Twp. (OUVC 3821). WASHINGTON Co. ne Sec. 3, Warren Twp. (OUVC 3295); Sec. 4, Warren Twp. (OUVC 2683); Marietta (OSM 620); Squaw Hollow (MC); Belpre Twp. (OSM 586); e Muskingum Twp. (OUVC 3354); Sec. 9, Lawrence Twp. (RB 91); Sec. 16, Ludlow Twp. (OUVC 3685); nw Sec. 13, Grandview Twp. (RB 102); Sec. 23, Dunham Twp. (OUVC 3770); Sec. 14, Watertown Twp. (OUVC 3771); Sec. 31, Wesley Twp. (OUVC 3772-73); Cutler, Fairfield Twp. (CMNH ZF1335) (fig. 10).

Insufficient comparative material from the east prevents our expressing any opinion on the validity of *rivicola* as a distinct race.

On April 12, 1957, 36 eggs were found attached to the bottom of a rock about one foot square. The rock was lying half under the water along the bank of a rock-strewn creek about

2½ miles southeast of Bainbridge, Ross Co. Several of these eggs were taken back to the laboratory, kept in pond water, and allowed to hatch. On April 24, the first eggs hatched and the larvae measured 11 mm. Other eggs were collected May 13, 1958, in a stream in Enderlin Forest, near Carbondale, Athens Co. Ten days later, they hatched.

Eurycea longicauda longicauda Green, Long-tailed Salamander. —ATHENS Co. Athens (OUVC 1553); Carbondale (OUVC 1555); Sec. 28, Canaan Twp. (OUVC 2177); Sec. 33, Canaan Twp. (OUVC 3936); Guysville (OUVC 2842); 7 mile se of Athens (OUVC 1402). GALLIA Co. Raccoon



FIGURE 10. The distribution of *Eurycea bislineata*.

Creek (OSM 885). HOCKING Co. Salt Creek, Benton Twp. (OUVC 1547); Cantwell Cliffs (OSM 677, 829); Neotoma (OSM 817); Clear Creek Valley (OSM 69, 91, 239); Sec. 24, Goodhope Twp. (RB 7). JACKSON Co. Sec. 14, Liberty Twp. (OUVC 2994); White's Gulch, Liberty Twp. (OSM 673); Jackson (OUVC 1187); Lake Jackson, Jefferson Twp. (OUVC 2399); Rock Run (OSM 833); near Byer, Washington Twp. (OSM 726). LAWRENCE Co. Hecla (OSM 838). MEIGS Co. Sec. 16, Bedford Twp. (OUVC 533); 2 mile e of Bashan (OSM 818); Pomeroy (OSM 433); Forked Run State Park, Olive Twp. (OUVC 3567-68). ROSS Co. near Tucson, Harrison Twp. (OSM A487); Chillicothe (Morse, 1904). SCIOTO Co. Harrison Twp. (OSM 384); Shawnee S.F. (DMD). VINTON Co. Knox Twp. (OSM 286). WASHINGTON Co. Squaw Hollow (MC); ne Sec. 26, Ludlow Twp. (OUVC 3691) (fig. 4).

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RELATIONSHIP BETWEEN TOTAL ALKALINITY, CONDUCTIVITY, ORIGINAL pH, AND BUFFER ACTION OF NATURAL WATER

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INTRODUCTION

In an effort to elucidate the properties of water related to the carbon supply for living organisms, the buffer action, conductivity, and total alkalinity of various natural waters were studied. Natural waters around Bowling Green, Ohio, were sampled at various times: Urschel's Quarry in Bowling Green being sampled most frequently; Klein's Well represented a soft ground water drawn directly from the ground by a pump; Poe Ditch was contaminated by sewage.

Dye (1944), using equilibrium equations of Moore (1939), produced nomographs for the relationship between pH, total alkalinity, and the three forms of carbon dioxide (free carbon dioxide, carbonate, and bicarbonate). However, Verduin (1956a) showed that these nomographs do not give a valid relationship between pH change and carbon dioxide change in natural water. The values obtained from the equilibrium equations do not agree with the actual data from Sandusky Bay, ocean water, and Lake Erie which Verduin (ibid.) has reported. Moberg et al. (1934) showed that their theoretical values do not completely agree with their computed values for ocean water. In the pH range from 6.0 to 8.0, observed linear increase of carbon dioxide content with decreasing pH is not in

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line with the theoretical values predicting an exponential increase in this region. Thus, other factors appear to influence the buffering capacity of natural waters.

METHODS

The samples were dipped from the surface of the water body in a 1,000 ml flask and returned to the laboratory, where the fresh samples were processed immediately. The aerated samples were allowed to reach equilibrium with the atmospheric CO_2 by standing at a depth of 2 cm in a large pan for at least 24 hr before processing.

A Barnstead Purity Meter (Model PM-3) was used to measure the electrical conductivity (Daniels et al., 1941). The meter was calibrated in megohms (Mohm) resistance and has a built-in temperature correction rheostat. The electrical conductance in mho of a 100-ml sample of distilled water was measured. The equipment was then rinsed thoroughly with distilled water. A sample of natural water was then diluted 1:50 with distilled water, mixed, and the conductivity measured. Then, 1:100 and 1:200 dilutions of the natural water were made and also measured for conductivity. The average of the three readings was computed to make the data more reliable.

A Beckman pH Meter (Model G) was used to support total alkalinity and buffering action titrations by methyl orange and phenolphthalein indicator methods (Kline, 1955; Bull, 1943). After three drops of phenolphthalein indicator had been added, 0.01 N H_2SO_4 or 0.02 N NaOH were titrated into the sample until the end point had been reached. Then, 1 ml of titrate at a time was added in excess until a total of 5 ml had been added. The total alkalinity was measured by titration to the visual end point with 0.01 N H_2SO_4 , using methyl orange indicator (Welch, 1948). The pH of the sample was measured before, during (in the case of phenolphthalein), and after titration.

Results of the phenolphthalein titration were graphed on arithmetic graph paper. From these buffer curves the micromoles of CO_2 per liter per pH unit were calculated for each four-tenths of a pH unit change. These data were then graphed against total alkalinity, original pH of the water, and conductivity.

Standard statistical computations were made (Snedecor, 1946). Since Klein's Well and Poe Ditch data were noticeably different from the natural water data, they were omitted from the statistical computations but were placed in the graphs for comparison.

RESULTS

The data were compared by means of graphs. Since the hydrogen ions and the hydroxyl groups react with an equal number of carboxyl groups, the micromoles of carbon dioxide change per unit of pH change can be computed from such a graph as follows:

$$\frac{\text{ml titrate} \times 10 \mu\text{mole CO}_2/\text{ml titrate}}{0.1 \times 0.4 \text{ pH unit}} = \mu\text{mole CO}_2/(\text{l} \cdot \text{pH unit})$$

Typical values for the buffer curve were 462.5 μmole of CO_2 (1 pH unit) for the pH range 7.0-7.4; 475 for the pH range 7.4-7.8; 500 for pH range 7.8-8.2; 450 for pH range 8.2-8.6; 700 for pH range 8.6-9.0; and 1025 for the pH range 9.0-9.4.

The conductivity graphed against the total alkalinity of the natural water samples (fig. 1) increased as the total alkalinity increased. The coefficient of correlation ($r = +0.72$) was highly significant, statistically ($P = <0.01$).

The original pH graphed against the buffer curve (fig. 2) showed a wide scatter with a correlation coefficient of +0.27. Similar graphs of original pH vs. total alkalinity and conductivity showed low correlations.

The total alkalinity graphed against the slope of the buffer curve in the pH range 8.2 to 8.6 showed that the buffering capacity of the water increased with

total alkalinity and has a correlation coefficient of $+0.76$ ($P=0.02$). All other pH ranges studied (7.0-9.4) showed similar positive, statistically significant correlations.

Conductivity graphed against the slope of the buffer curve in pH range 7.4 to 7.8 has a correlation coefficient of $+0.32$, and this graph was representative of the other pH ranges. The product of total alkalinity and conductivity was graphed against the slope of the buffer curve (fig. 3) to learn whether this product

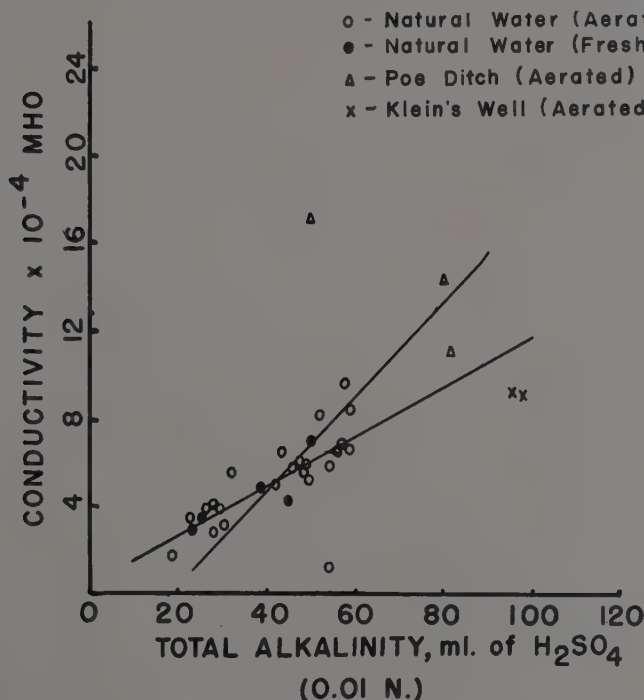


FIGURE 1. Relation of conductivity to total alkalinity.

would show a higher correlation than each property by itself. There is a wide variation to the data; however, a correlation coefficient of $+0.52$ ($P=0.04$) was found. The correlation coefficient is greater than conductivity graphed against the slope, and less than total alkalinity graphed against the slope of the buffer curve. This then showed that factors other than the forms of carbon dioxide affect buffer action, but these have not been investigated. A summary of the statistical analyses appears in table 1.

DISCUSSION AND CONCLUSIONS

Variation in limnological data is normal and to be expected. Powers (1939) found a range of over one pH unit in the blood pH and aerated serum within the same species of fish. The relation of Secchi disc readings to the depth associated with one percent of surface light presents also a scatter of the data (Verduin, 1956b). The photosynthetic rate at optimal light intensity vs. respiration rate demonstrates a wide range with values ranging from 0.25 to 14 $\mu\text{mole of CO}_2/10\mu\text{l}\cdot\text{hr}$ (Verduin, 1956a). In terrestrial biological data variation is also observed,

as in a measure of the wood mouse (*Peromyscus leucopus*) population (Jackson, 1952). Environmental variations do occur, and the researcher must accept them.

In graphs presented by Moberg et al. (1934) variations are as great or greater than the variation observed for the present data, and portions of their observed data differ markedly from their theoretical values. This is especially noticeable below the pH range of 7.5. Their theoretical equation apparently does not account for all of the dissolved ions in the water. Even after introducing a correction factor for boron into their equation, they observed values still at wide

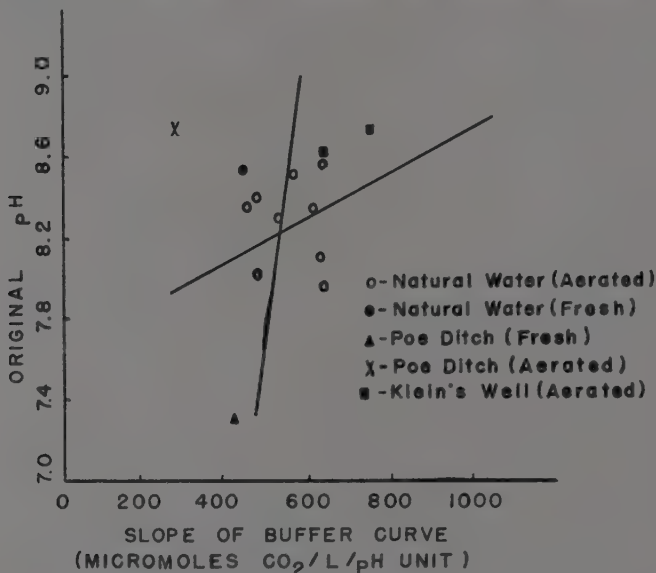


FIGURE 2. Relation of original pH to slope of buffer curve (pH range 7.4-7.8).

variance with their calculated values at low pH. Since the chemical conditions are different for each body, and are constantly varying within each body, wide variance in the data may be influenced by varying chemical conditions of the water.

The literature discusses the effects of many chemicals present in water. While many are present in only trace amounts, they do influence biological processes. Many are essential for the growth of plants and are present in ionic form, thus contributing to the total conductivity. In supporting plant life, they indirectly affect respiration and photosynthesis and the use of carbonates and bicarbonates. Mollusca, *Elodea*, and *Vallisneria* precipitate calcium carbonate as marl and shells. Since calcium carbonate is insoluble, a decrease in the total alkalinity, conductivity, and buffer capacity of the water will result. Chemical conditions of such water therefore vary greatly and thus affect variations in the buffer curve, total alkalinity, pH, and conductivity.

The aerated and fresh samples did not differ significantly from natural water. Since the water was always collected from the surface, it is likely that the fresh sample was already in equilibrium with the atmospheric gases.

The phenolphthalein and methyl orange indicators are used widely in limnological studies. These indicators are assumed to indicate presence of carbonates

and the absence of carbonic acid (Welch, 1948; Verduin, 1959). However, in this study no significant differences were found between the aerated and fresh samples. Thus, an aerated sample must contain some carbonic acid while the carbonates must be practically absent. Carbon dioxide is continually entering the water, converting to carbonic acid, and changing the carbon dioxide into bicarbonates. Thus, phenolphthalein alkalinity measures the hydroxyl ions rather than the carbonates, as one is so often led to believe.

Poe Ditch and Klein's Well differ markedly from natural water. Samples from these two sites had a much higher electrical conductance and total alkalinity,

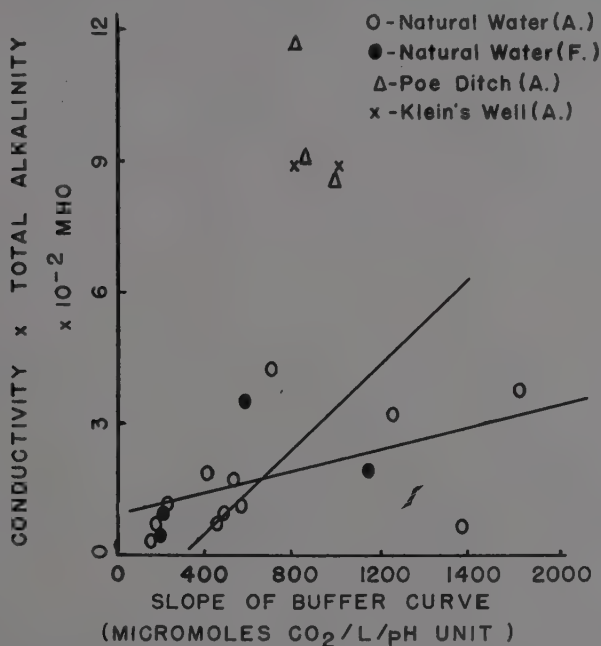


FIGURE 3. Relation of product of conductivity and total alkalinity to slope of buffer curve (pH range 8.2-8.6).

but the slopes of the buffer curves were intermediate to those for natural water. The product of total alkalinity and conductivity, as might be expected, further separated the ditch and well samples from the natural waters.

Factors other than the various forms of carbon dioxide affect buffering action. Since the coefficient of correlation of total alkalinity vs. the slope of the buffer curve ($r = +0.52$) is intermediate between conductivity vs. slope ($r = +0.32$) and total alkalinity vs. slope ($r = +0.76$), other unmeasured ions affect the buffer curve. When the product of conductivity and total alkalinity was graphed against slope, the unmeasured ions were partially masked; when total alkalinity was graphed against slope, the unmeasured ions were entirely absent, thereby, resulting in a high coefficient of correlation.

However, these studies have not explained the wide variation in the slopes of the buffer curves. Verduin (1956a) has shown differences in the buffer curves for a standard sodium bicarbonate solution, ocean water, Sandusky Bay water, and Lake Erie water; and the present study revealed similar results.

TABLE 1
Summary of statistical analyses*

	N	b_{xy}	b_{yx}	t_b	P_b	r	t_r	P_r
pH vs. Slope	11	0.0011	64.24	0.85	<0.01	+0.27	2.63	0.02
Conductivity vs. total alkalinity	34	10.99	0.047	5.36	<0.01	+0.72	5.30	<0.1
Conductivity vs. slope	14	0.47	0.22	0.96	0.34	+0.32	0.99	0.30
Total alkalinity vs. slope	21	0.021	30.34	1.14	0.23	+0.76	2.56	0.02
Total alkalinity X conductivity vs. slope	16	12.74	0.21	1.73	0.11	+0.52	2.25	0.04

*KEY

N=Number of pairs of values used for each analysis.

 b_{xy} =Slope of line for x on y. b_{yx} =Slope of line for y on x. $t_b = b/\Sigma b$. P_b =Probability. r = Correlation coefficient = $\Sigma xy / \sqrt{(\Sigma x^2)(\Sigma y^2)}$ $t_r = r\sqrt{(n-2)/(1-r^2)}$ P_r = Probability of correlation coefficient.

SUMMARY

1. Factors other than the three forms of carbon dioxide affect buffering capacity.
2. Buffering action was least efficient between the pH range 8.2 to 8.6.
3. Ions other than the forms of carbon dioxide were found to affect the buffer curve. These are unmeasured at present.
4. Phenolphthalein alkalinity measures the hydroxyl ions rather than carbonate ions alone.
5. Apparently the pH of the natural waters is relatively independent of total alkalinity, conductivity, and buffer capacity of those waters.

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A KEY TO THE *EMPOASCA SOLANA* COMPLEX WITH DESCRIPTIONS OF NEW SPECIES¹

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In the extremely useful synopsis of the complex by Young (1953a), 12 species were recognized and since that time Young (1953b) has added one more described species. Examination of large collections of *Empoasca* from chiefly the circum-Caribbean area in connection with *Empoasca fabae* studies has disclosed the existence of an additional 11 species, bringing to 24 the total number of known species in the complex. No species are known outside the western hemisphere. In this paper a key is given to these 24 species; newly discovered species are described and their relationships indicated. Unless otherwise indicated, types are deposited in the collection of the Illinois Natural History Survey with paratypes where available deposited in the D. M. DeLong collection and the U. S. National Museum.

We wish to take this opportunity to express our deep gratitude to Dr. G. C. Decker, our co-investigator in Project NC-29, for his untiring efforts in collecting and procuring study material in this group. Dr. J. E. Porter and Dr. Irving Fox, U. S. Public Health Service, and Captain R. M. Altman, U. S. Navy, have also been extremely generous of their time in sending us residues from light trap collections containing *Empoasca* specimens.

In attempting to determine the species limits in this complex, we encountered the usual difficulties as regards the status of several unique specimens and short series. In this we were aided greatly by the very large series of specimens available for *E. solana*, *manda*, *stevensi*, and four or five other species in the complex. In these species the discernible variation was of much smaller magnitude than that occurring between several of the unique specimens which were ultimately considered as valid species. It is somewhat peculiar that the species represented by small series are almost invariably much better differentiated morphologically than some of the commoner species which apparently occur in great numbers.

GENERAL DESCRIPTION OF COMPLEX

All the species in this complex are remarkably similar in general characteristics and the following description applies to all the new and old species mentioned or described in this paper, except where noted: Length 3.5 mm; color in life pale green with inconspicuous translucent markings on the head and thorax; male abdomen with long apodemes 2S which may be either slightly truncate or rounded at apex and which extend almost three segments into the abdomen; apodemes 3T very small and represented by either a linear structure or a very slight elliptic widening. Hook of tenth segment moderately large but forming a rounded plate without points or spurs, only slightly sclerotized and inconspicuous (DeLong 1931, fig. 10). Aedeagus having a pair of spines situated on or below the shaft.

In most species the shaft has an internal clear area which in transmitted light is sharply defined; this area, here called the aedeagal sinus, usually ends dorsally in a tapered point and in many species has a diagnostic shape.

Key to Species - Males

1. Apodemes 3T larger and as long as apodemes 2S, fig. 15. Widespread.....
thela Davidson and DeLong
Apodemes 3T no longer than in fig. 14, apodemes 2S much larger, fig. 16. 2

¹A contribution from North Central Regional Project NC-29.

2. Aedeagal spines short and situated far out on shaft (Young 1953a, fig. 1B). Haiti.....
canavalia DeLong 3
 Aedeagal spines arising at or below base of shaft..... 3
3. Both shaft and aedeagal spines short and wide, fig. 3A. Panama..... *canthella* n. sp.
 Shaft and aedeagal spines much longer, figs. 5, 6..... 4
4. Shaft elongate and slender, as in figs. 1A, 13A..... 5
 Shaft more robust..... 9
5. Aedeagal spines appressed to sides of shaft (Young 1953a, fig. 8B); brachones swollen.
 Tropical..... *wolcottii* Young
 Aedeagal spines situated beneath shaft, fig. 13A; brachones not swollen..... 6
6. Aedeagal spines fused for about three-fourths of their length, fig. 13D. Mexico.....
caldwelli Davidson and DeLong 7
 Aedeagal spines separate..... 7
7. Tip of aedeagus forming a narrow, sharp point (Young 1957, fig. 27C). Bolivia.....
hyalina (Osborn) 8
 Tip of aedeagus as thick as in fig. 1A..... 8
8. Posterior aspect of brachone with tip smoothly sinuate. Widespread.....
plebeia DeLong and Davidson
 Posterior aspect of brachone with tip having a fairly abrupt, steplike notch, fig. 1C.
 Southwestern U. S., Mexico..... *ophiodera* n. sp.
9. Shaft with a well-marked ventral corner and aedeagal spines extending at most only a
 trifle beyond this corner (Young 1953a, fig. 3B)..... 10
 Shaft without a well-marked ventral corner and aedeagal spines longer, figs. 4A, 12A..... 12
10. Aedeagal spines running close to ventral edge of shaft; aedeagal sinus trianguloid
 (Young 1953a, fig. 3B). Widespread..... *solana* DeLong
 Aedeagal spines arising below base of shaft, sometimes angling or curving to touch ventral
 corner of shaft..... 11
11. Brachone having a broad angulate shoulder just below tip (Young 1953a, fig. 4D);
 aedeagal sinus fairly shallow and arcuate. U. S., West Indies.....
manda Davidson and DeLong
 Brachone without subapical shoulder but sinuate as a whole; aedeagal sinus and ventral
 corner of shaft produced postero-ventrally into an acute angulation (Young 1953a,
 fig. 2B). Widespread..... *stevensi* Young
12. One aedeagal spine large and thick, the other much shorter, either wide and sharp, figs.
 6A, 7A, narrow, or obsolescent, figs. 8A, D..... 13
 Both aedeagal spines of about the same length..... 15
13. Shorter aedeagal spine as long as in figs. 6A and 7A..... 14
 Shorter aedeagal spine shorter and rounded, figs. 8A, D, or represented only by an in-
 distinct swelling. Florida, Central America..... *deckeri* n. sp.
14. Shorter aedeagal spine slender and projecting outside orbit of larger spine (Young 1953b,
 fig. 7B). Argentina..... *trilabulata* Young
 Shorter aedeagal spine stout and projecting within orbit of larger spine, figs. 6A, 7A.
 Puerto Rico, Panama..... *teneris* n. sp.
15. Aedeagal spines contiguous with edge of shaft, figs. 2A, 5A, 9A, 10A..... 16
 Aedeagal spines angling away from shaft then curving back to touch or overlap it, figs.
 4A, 11A, 12A..... 20
16. Aedeagal sinus fairly short and stout, not extending beyond tips of aedeagal spines,
 fig. 2A. Central America..... *cristella* n. sp.
 Aedeagal sinus longer..... 17
17. Apex of aedeagus curved back on its base very gently, fig. 5A. Puerto Rico..... *foxi* n. sp.
 Apex of aedeagus sharply angled back over its base, figs. 9A, 10A..... 18
18. Aedeagal spines sinuate, aedeagal sinus pinched almost to a thread below apex (Young
 1953a, fig. 7B). Widespread subtropical..... *tamiana* Davidson and DeLong
 Aedeagal spines curved, fig. 9A..... 19

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|-----|--|------------------------|-------------------------------------|
| 19. | Shaft short, aedeagal sinus narrow and tapering at apex, fig. 9A. | Panama..... | <i>alceda</i> n. sp. |
| | Shaft long and stout, aedeagal sinus wide almost to tip, fig. 10A. | Peru..... | <i>robachi</i> n. sp. |
| 20. | Shaft only slightly curved, aedeagal sinus short, narrow, and gently arcuate (Young 1953a, fig. 5B). | Southwestern U. S..... | <i>dilitara</i> DeLong and Davidson |
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| 22. | Aedeagal sinus having an enlarged, rounded apex, fig. 12A. | Mexico..... | <i>tecpatana</i> n. sp. |
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| 23. | Shaft short and stocky, its tip wide, fig. 4A. | Honduras..... | <i>lanceiba</i> n. sp. |
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Descriptions of New Species

Empoasca canthella n. sp.

Male.—Size, color, and general structure typical for the complex. Brachone (fig. 3*B*, *C*) with lateral aspect swollen toward base of apex, the apex itself narrow and nearly straight; with posterior aspect having a broad lateral shoulder at base of apical portion, the apical portion itself narrow and sinuate. Aedeagus (fig. 3*A*) with shaft small, somewhat irregular, with a short, sharp sinus clearly indicated; aedeagal spines short and blunt, arising at the base of shaft but projecting almost straight out.

Holotype male.—Balboa, C. Z., Panama, July 13, 1946, A. O. Meyer.

Paratype.—Farfan, C. Z., Sept. 15, 1955, R. M. Altman, light trap, 1 male.

The shape of the aedeagus of this species differs remarkably from any other described type in the complex, in both the low shaft and the very short spines. When the first specimen was encountered, we thought it might be a freak but the second specimen agrees so perfectly with the first that there seems to be a good chance that this is a distinctive species. The short and distinct sinus is reminiscent of *solana* and the swollen brachone is reminiscent of *manda*; on the basis of these two hints it is probable that this species represents an aberrant offshoot which arose from the *solana-manda* section of the complex.

Empoasca laceiba n. sp.

Male.—Size, color, and general structure typical for the complex. Brachone (fig. 4B, C) with lateral aspect only slightly swollen before apex, the apex slightly curved ventrad; and with posterior aspect more slender, again only slightly swollen, the apex narrow and very slightly sinuate. Aedeagus (fig. 4A) with sinuate base and with a very broad shaft which is angled fairly sharply back over the base; aedeagal sinus narrow, long at the base and also bent sharply and ending in a narrow point; aedeagal spines long, arising below main body of shaft and curving to overlap ventral corner of shaft, one of the spines wider than the other.

Holotype male.—Laceiba, Honduras, May 28, 1949, E. C. Becker, at light.

This species is closest in general structure to *stevensi*, from which it differs in the longer aedeagal spines and the longer basal part of the aedeagal sinus. In these two characters *laceiba* is strikingly similar to *olivata*, but from *olivata* it differs in the pronounced ventral corner of the shaft and the short and extremely stubby apex of the shaft, both characters distinctive for *stevensi*. This unusual combination of characters distinctive to widely separated phyletic lines may indicate that *laceiba* is of hybrid origin.

Empoasca cristella n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone (fig. 2*B*, *C*) with lateral aspect narrow and very slightly sinuate, the apical portion short and merging imperceptibly with the long slender base; with the posterior aspect also narrow, the apical portion curved and its mesal margin markedly concave. Aedeagus (fig. 2*A*) with the base fairly straight and the shaft broad, fairly short, and very slightly curved anteriorly; aedeagal sinus well marked, unusually broad, nearly touching the ventral margin and ending abruptly

in a short, sharp point; aedeagal spines appressed to ventral margin of shaft, extending above the level of the apex of the sinus, both spines fairly slender.

Holotype male.—Laceiba, Honduras, May 28, 1949, E. C. Becker, at light.

Paratype.—Farfan, C. Z., Panama, June 4, 1957, R. M. Altman, 1 male.

This species approaches only *dilitara* in the shape of the shaft but is readily distinguished from that species by the very wide aedeagal sinus and the longer aedeagal spines appressed to the shaft; in *dilitara* the aedeagal sinus is narrow and almost hornlike and the aedeagal spines curve well below the ventral margin of the shaft. There is also a marked resemblance between the aedeagus of *cristella* and that of *foxi* but in *foxi* the apex of the shaft is markedly curved anteriorly and the sinus, which is not well defined basally, extends well into the apex of the shaft. These comparisons indicate that *cristella* probably arose from the base of the *dilitara* line and is a primitive member of the complex.

Empoasca foxi n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone with lateral aspect narrow and even, the apical portion marked by a sinuate dark area, as in figure 8B; the posterior aspect also straight and narrow, the apex much like the lateral aspect. Aedeagus (fig. 5) with base slightly curved and with shaft fairly broad, its apex curved back anteriorly over the base; aedeagal sinus indistinct at base, its apex reaching well into the apical portion of the shaft and its posterior side near the posterior margin of the shaft; aedeagal spines slightly unequal, their tips curved anteriorly, both of the spines following fairly closely the margin of the shaft.

Holotype male.—Isla Verde, near San Juan, Puerto Rico, October-November, 1957, Irving Fox, at light.

Paratypes.—Same data as for holotype but July, 1 male; same but June, 1958, 2 males.

This species, undoubtedly a primitive one, is most similar to *cristella* but differs from it in the curved apex of the shaft and the much longer aedeagal sinus. This species was undoubtedly recorded from Puerto Rico by Caldwell and Martorell 1952 under the name *canavalia*, as evidenced by their remark that in some of their specimens the aedeagal spines are subequal. The holotype of *canavalia*, however, is unusual and quite different in its extremely reduced aedeagal spines, illustrated by Young (1953a, fig. 1B). The only additional specimen we have seen agreeing with the holotype of *canavalia* is a male collected by Capt. R. M. Altman, Dec. 11-18, 1956, at Ft. Kobbe, Canal Zone, Panama.

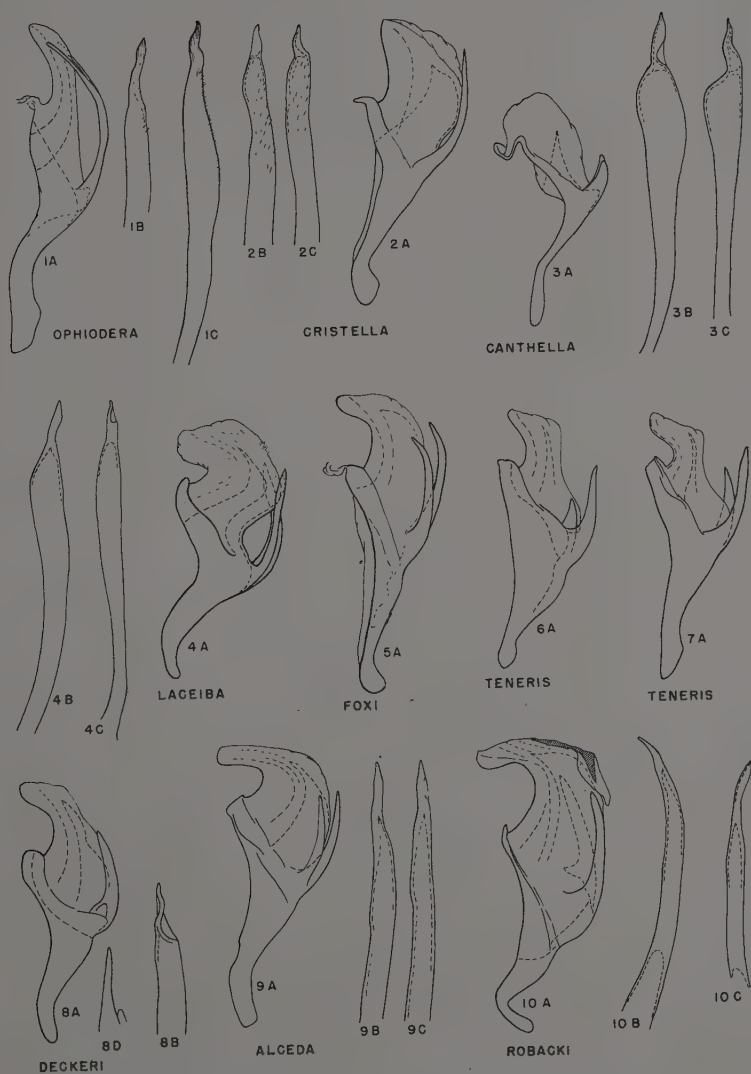
Empoasca deckeri n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone very long and slender (fig. 8B) the lateral aspect parallel-sided and ending in an apical portion which appears incised ventrally in steplike fashion; this steplike appearance, however, is due to a narrow sinuate darkening mesad of which there is an inconspicuous, poorly sclerotized area, so that the apex of the brachone is actually triangular. Aedeagus (fig. 8A) with base slightly sinuate; shaft broad at base, not very long but narrowed and slightly constricted below the apex, the whole curved markedly anteriorly over its base; aedeagal sinus poorly defined at the base and narrow and elongate apically; aedeagal spines extremely dissimilar, consisting of a long stout one appressed to margin of shaft, and a minute rounded stub contained within the silhouette of the base of the larger spine, this small one sometimes represented only by a darkened spot at the base of the larger spine. In some specimens the large spine is the right one and the small one the left one (fig. 8D) and in other specimens it is the reverse.

Holotype male.—Miami, Florida, Feb. 19, 1957, G. C. Decker, on purple morning glory.

Paratypes.—70 males from the following localities: Canal Zone, Panama: Farfan, Fort Cobbe, Curundu, Cocoli; Honduras: Laceiba, 12 k. west of Olanchito; Florida: Coral Gables, Key Largo, Miami, Key West, Homestead, Dade County; Nicaragua: Managua.

This species and the next are most closely related to *foxi*, from which *deckeri* may be separated by the extreme reduction of the smaller aedeagal spine and by the constricted apical portion of the shaft. From the following species, *teneris*, *deckeri* may be differentiated by the fact that the small aedeagal spine in lateral view is always contained within and obscured by the extreme base of the larger spine.



FIGURES 1-10. Male genitalia of species in the *Empoasca solana* complex. A--lateral aspect of aedeagus; B--lateral aspect of brachone; C--posterior aspect of brachone; D--posterior aspect of aedeagal spines.

Empoasca teneris n. sp.

Male.—Size, color, and general structure as described above for the complex. Male genitalia extremely similar to those of *deckeri*. Brachone long and parallel-sided, exactly like those of *deckeri* (see fig. 8B). Aedeagus (fig. 6 and 7) with nearly straight base; shaft broad at base and tapering to constricted central portion, beyond which it flares out again, the apex curved anteriorly back of its base; the posterior and ventral portion of the shaft is composed of rather delicate membranes which are frequently difficult to detect; aedeagal sinus with basal portion indistinct, its apical portion narrow and curving to a pointed apex. Aedeagal spines markedly asymmetrical, the longer one stout and angled dorsally away from the shaft; the shorter one, also stout and sharply pointed, projecting between the longer one and the base of the shaft as seen from lateral view. The length of the shorter spine varies considerably between the two extremes illustrated in figures 6 and 7. As in *deckeri*, both right and left-handed forms occur.

Holotype male.—Isla Verde near San Juan, Puerto Rico, July, 1957, Irving Fox, at light.

Paratypes.—Panama, Canal Zone: Fort Cobbe, January 3–7, 1957, R. M. Altman, at light, 1 male; and 140 males from various dates collected at the holotype locality.

This species is a close relative of *deckeri*, sharing with it the characteristically constricted shaft and the remarkable asymmetry of the aedeagal spines. From *deckeri*, *teneris* can be differentiated by the outwardly directed larger aedeagal spine and the longer and sharper, small and inner aedeagal spine.

This is the species described and illustrated from Puerto Rico by Caldwell and Martorell (1952, plate 53) under the name *canavalia* (see note concerning type of *canavalia* in description of *foxi* above).

Empoasca alceda n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone (fig. 9B, C) with lateral aspect elongate, parallel-sided, and slender, the apical portion represented by a more slender and pointed straight terminus; the posterior aspect also narrow and slender although slightly irregular, the apical portion narrow and pointed. Aedeagus (fig. 9A) with base sinuate; shaft with basal portion deep and apex angled anteriorly back over base to form an almost rectangular portion; aedeagal sinus wide at base, its posterior and ventral margin merging with the margin of the shaft, its apex produced into a moderately long, narrow, and sharp tip. Aedeagal spines moderately short and slender, appressed to the shaft for much of their length.

Holotype male.—Camp Pina, Canal Zone, Panama, July 5, 1955, R. M. Altman, at light.

Paratypes.—Canal Zone Panama: Same data as for holotype, 1 male; Cocoi, September 16, 1955, R. M. Altman, at light, 1 male; Fort Gulick, February 8, 1957, R. M. Altman, at light, 1 male.

The strongly reflexed apex of the shaft indicates a relationship between this species and *olivatula* and its allies; from this group of species *alceda* differs in having shorter and thinner aedeagal spines. Other characters of the aedeagus indicate a similarity with *foxi*, from which *alceda* differs in the strongly reflexed shaft and the narrow, pointed apex of the aedeagal sinus. This combination of characters suggests strongly that *alceda* represents an offshoot from a primitive stock of the complex and that this offshoot was also the base of the more specialized species such as *olivatula* in which the reflexed shaft is prominent.

Empoasca robacki n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone (fig. 10B, C) with lateral aspect curved gently dorsad, the basal portion narrowing imperceptibly into an elongate, slightly sinuate, sharply pointed apex, with posterior aspect also narrow and parallel-sided, narrowing more abruptly into an apical portion which is parallel-sided and curved slightly mesad. Aedeagus (fig. 10A) with base small and curved; shaft large and massive, its apex reflexed sharply and its postero-dorsal portion having unusually developed folds of membrane; aedeagal sinus indistinct at base but the apical portion broad and extending to the top of the shaft. Aedeagal spines large and massive, contiguous with or overlapping the shaft.

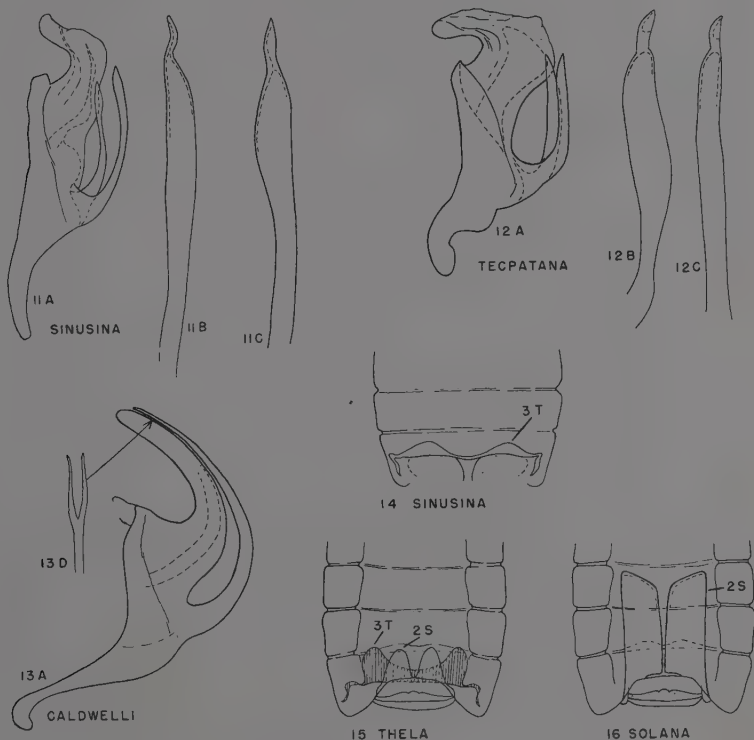
Holotype male.—Tinga Maria, Peru, October 6, 1955, S. Roback, at light.

This species is most closely related to *alceda* but differs from it in the unusually large and

massive shaft and the broader aedeagal spines. The more massive shaft is somewhat suggestive of the two following species but in these the aedeagal spines curve far below the body of the shaft.

Empoasca sinusina n. sp.

Male.—Size, color, and general structure as described above for the complex. Apodemes 3T (fig. 14) produced into fairly wide crescentic arcs. Brachone (fig. 11B, C) with lateral aspect fairly narrow, tapering to a fairly long apical portion which is curved and narrow; and with the



FIGURES 11-13. Male genitalia of species in the *Empoasca solana* complex. A—lateral aspect of aedeagus; B—lateral aspect of brachone; C—posterior aspect of brachone; D—posterior aspect of apex of aedeagal spines.

FIGURES 14-16. Base of male abdomen of species in the *Empoasca solana* complex. 14—dorsal aspect of apodemes 3T; 15—ventral aspect of apodemes 2S and 3T; 16—ventral aspect of apodemes 2S.

posterior aspect somewhat swollen toward the apex, this swollen portion narrowing gradually to the base of the apical portion which is slightly constricted below middle and pointed at the tip. Aedeagus (fig. 11A) with nearly straight base; shaft narrowing abruptly just beyond base, fairly long, and with the apex reflexed markedly over the base; the posterior margin of the shaft is membranous and sinuate; aedeagal sinus extremely narrow, sinuate, and varying in thickness, ending

in a sharp, narrow point. Aedeagal spines elongate, one slightly shorter and more slender than the other, both curving away from the shaft and then curving back toward it.

Holotype male.—Navy Ordnance Headquarters, Canal Zone, Panama, June 1, 1955, R. M. Altman, at light.

This species is most closely related to *olivatula* but differs from it in the much narrowed shaft and aedeagal sinus. The species undoubtedly represents an offshoot of the *olivatula* line.

Empoasca tecpatana n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone (fig. 12B, C) with lateral aspect unusually wide, tapering to a much narrower and somewhat finger-like apical portion; posterior aspect narrow and parallel-sided, nearly straight, narrowing to the apical portion which is somewhat finger-like from this aspect also. Aedeagus (fig. 12A) with short curved base, the posterior margin having a produced rounded lobe just below the bases of the aedeagal spines; shaft moderately wide, its apex reflexed sharply over its base, its posterior margin concave; aedeagal sinus well marked for its entire length, its apical portion expanded and ending in a broadly rounded lobe. Aedeagal spines nearly equal in length and size, first angling away from and beneath body of shaft, then curving into straight portions which touch the posterior corner of the shaft.

Holotype male.—Tecpatan, Chiapas, Mexico, August 30, 1946, A. Dampf, at light.

This species is most closely related to *olivatula*, differing from it in the nearly equal aedeagal spines (in *olivatula* one is about the width of the other) and in the wide and rounded aedeagal sinus. In *tecpatana* the lateral aspect of the brachone is much wider than in *olivatula*. It seems highly likely that *tecpatana* represents a specialized offshoot of the *olivatula* line. It is of further interest that the species *tamiama* which has a peculiar conformation of the aedeagus (illustrated by Caldwell and Martorell, 1952, plate 53, as *everaformis*) is very likely a specialized offshoot of *tecpatana*. If one visualizes starting with an aedeagus such as that in *tecpatana*, squeezing the sinus together until it is divided in the middle, twisting the aedeagal spines so that they lie against the shaft, and enlarging the bulge on the base below the aedeagal spines, the result is essentially that of the structure found in *tamiama*.

Empoasca ophioidera n. sp.

Male.—Size, color, and general structure as described above for the complex. Brachone (fig. 1B, C) with lateral aspect long, slender, and parallel-sided, tapering to a more slender apex; posterior aspect with ventral portion slightly enlarged and with apex sinuate and having a somewhat steplike notch on the mesal margin. Aedeagus (fig. 1A) with base sinuate and narrow; shaft also narrow and elongate, slightly curved anteriorly and with a simple type of gonopore, without any indication of a sinus. Aedeagal spines long and slender, curving away from the shaft at base but recurving to overlap the tip of the shaft.

Holotype male.—Sabino Canyon, Tucson, Arizona, October 13, 1957, Bibby and Benny, on *Salix*.

Paratypes.—Arizona: Tortilla Flat, October 27, 1957, F. F. Bibby, on *Baccharis glutinosa*. Mexico: Cotaxtla, Veracruz, September 10, 1957, W. W. Gibson, on *Sesamum*.

This species is most closely related to *plebeia* from which it differs in the shorter, thicker, and incised apex of the brachone; in *plebeia* the apex of the brachone is more slender, longer, and smoothly sinuate.

EVOLUTION OF THE COMPLEX

In surveying the various species described in the *solana* complex, it is apparent that all of the species form a closely knit complex sufficiently distinct from all other described species in the genus that the complex is monophyletic. Deducing the ancestral form from which the complex arose, however, is fraught with some difficulty. In the present attempt to arrange the species in a logical evolutionary sequence, the most satisfactory and plausible arrangement has been achieved by considering that forms such as *ophioidera* (fig. 1) or *foxi* (fig. 5) are the closest approach to the ancestor of the group. In these the aedeagal spines are long and

equal, the aedeagal sinus is poorly defined, and the shaft is not markedly angulate either in the form of a recurving apex or a postero-ventral shoulder. If the ancestor of *ophiodera* ever did have an aedeagal sinus, then this line probably arose early in the evolution of the complex and forms with *plebeia* a separate branch. If this is the case, the ancestral form of the complex also had aedeagal spines which bowed out below the shaft so that the hypothetical ancestor for the rest of the complex would probably have represented a combination of the shaft of *foxi* (fig. 5) and the aedeagal spines of *dilitara*, much like those in figure 4A. From such an ancestor two main lines probably arose, one characterized by a shortening of the aedeagal sinus and the development of a pronounced postero-ventral shoulder on the shaft; *cristella* (fig. 2) and *dilitara* may represent early stages in this lineage and *stevensi* a later stage. The development of the postero-ventral shoulder of the shaft was accompanied by the shortening of the aedeagal spines. To this entire branch we are assigning *cristella*, *dilitara*, *solana*, *canthella*, *manda*, *laceiba*, and *stevensi*. The species *canthella* (fig. 3) would appear to be an example of extreme reduction of the aedeagal parts. The species *laceiba* (fig. 4) is the only misfit and it will be discussed later. The other main branch of the genus contains a group of nine species in which the apex of the shaft bent back over its base and in extreme cases became almost angular in this relationship. The first step in this development is represented by *foxi* (fig. 5) *teneris* (figs. 6, 7) *deckeri* (fig. 8) and perhaps *tritabulata*, which have the apex of the shaft only gradually reflexed. The next step is probably best represented by *alceda* (fig. 9) and *robacki* (fig. 10) in which the apex of the shaft is much more angularly reflexed. This characteristic is present also in *olivatula*, *sinusina* (fig. 11), *tecpatana* (fig. 12), and *tamiamia*, indicating that these species arose from an *alceda*-like progenitor. This leaves unplaced the three species *hyalina*, *caldwelli* (fig. 13) and *thela*, in which the shaft is long, extremely slender, and also curved markedly anteriorly though not angled as in the *alceda* complex. It is probable that these three species form a monophyletic branch which could have arisen either from the *plebeia* line or even possibly from the base of the *alceda* line. The species *canavalia* is impossible to place with any degree of certainty because it is so dissimilar from other species.

The species *laceiba* introduces an interesting point of speculation. In the *solana* branch it would appear as if the development of the postero-ventral corner of the shaft had gradually become more and more pronounced and as if the sinus had been pulled down into the corner of the shaft. The shape of the aedeagus of *stevensi* is further suggestive that the entire shaft has been distorted postero-ventrally. If this is the case then the angulation produced on the anterior margin of the shaft is the result of this pull and presumably this same trend has brought about the shape of the shaft in *laceiba*. In *laceiba*, however, the aedeagal spines are elongate, more as in *tecpatana* or *olivatula*. There is here a suggestion that *laceiba* may be the result of hybridization between a form somewhat like *stevensi* and one somewhat like *olivatula*.

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AN EXPERIMENT IN BIOLOGY TEACHING

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During the first semester of 1956, an educational experiment was carried on in the Biology Department of Bowling Green State University. The experiment included all students registered in the two general biology courses (about 750) and all except one of the ten staff members in the department. It consisted in giving all students open book objective type examinations at three week intervals during the course. The examinations were all prepared by the same author, and students were informed at the beginning of the course that these examinations would be given. They were also supplied with a list of reading assignments covering the entire semester, and were informed that they would be permitted to use their books during the examinations.

The examination questions were all of the multiple choice type, containing five choices for each question. For example: The elements hydrogen, carbon, oxygen, and nitrogen account for about—

(a) 10% (b) 26% (c) 52% (d) 65% (e) 96%

of the material in protoplasm. The information needed to answer this question correctly is contained in a table in the text, the student only needs to add up the percentages of the elements mentioned to obtain the answer of 96 percent. More than half of the questions on each examination were of this general type, requiring only a reasonable familiarity with the textbook, and an ability to read comprehensively. Each test contained twenty questions and the students were given a fifty minute period for taking the test.

In addition to the simple type of question described above, each test contained several questions which required more refined analysis. For example: Figure 4:9, page 96, contains information indicating that *Chlamydomonas* is:

(a) motile (b) capable of photosynthesis (c) capable of storing food (d) all of the above (e) none of the above. The figure referred to shows this small aquatic plant and identifies its flagella (which impart motion), its chloroplast (which is an agent of photosynthesis), and its pyrenoid (a food storage structure). In other parts of the text the function of these structures is described so the student must recognize these functions when he examines the figure in question. He must also recognize that the only acceptable answer to this question is "(d) all of the above," because no credit is given for selecting one of the "correct" statements but failing to recognize that the other two are also correct. Questions requiring such more refined analysis comprised about one-third of the twenty questions on each test.

The two general biology courses differed considerably. One was a one-semester nonlaboratory course in which the sections contained about 45 students and were met in three one-hour meetings per week. The other was a two-semester laboratory course in which sections did not contain more than thirty students and the students attended two one-hour meetings and two two-hour laboratories per week. Moreover, the nonlaboratory course was taken by students who did not intend to take additional biology courses, while the laboratory course was taken by students who were planning more intensive study in biology. Most biologists are agreed that a nonlaboratory course is inferior to a laboratory course, and that a one-semester "terminal" course provides only a superficial survey of biology. It is interesting to compare the performance of the two groups of students in these courses. Data gathered in this experiment are shown in table 1. The average grade in the nonlaboratory course was 64 percent, and the average grade in the laboratory course was 58 percent. Thus, it appears that the addi-

tional time spent in the laboratory course, and the presumably greater motivation for careful study on the part of students who intend to continue in biology, did not enable these students to understand the textbook and the examination questions more clearly than did the students who were taking the nonlaboratory course.

In the laboratory course some double sections were met in a lecture room for the one-hour meetings and these sections were met in laboratory by graduate assistants who were completely responsible for the laboratory instruction. A total of eight sections was handled in this fashion. In the other eleven sections both lecture and lab were met by the same instructor. A comparison of grades in these groups did not show a significant difference. The students who attended "single" lecture sections and were taught by the same instructor in laboratory did not show a clearer understanding of the textbook and the examination questions, than did the students who had to attend lecture in larger groups and had graduate students for laboratory instruction.

Moreover, the methods of instruction varied considerably from one instructor to another. Some instructors lectured extensively, others used primarily a recitation-discussion procedure. Some instructors had many years of experience,

TABLE I
Comparison of grades (percentage correct) of various groups

	Avg.	Number of sections in sample
Nonlaboratory course	64 (92-30)*	5
Laboratory course	58 (86-31)	17
Large lecture section with graduate students responsible for labora- tory instruction	58 (86-28)	8
Small lecture sections. Laboratory and lecture by same instructor	58 (86-31)	9
Sections of experienced instructors	58 (86-30)	10
Sections of first year instructors	59 (86-29)	7

*The values in parentheses indicate the average range within sections.

others were in their first year of employment. A comparison of the performance of students subjected to these different methods and different degrees of instructor's experience did not reveal consistent differences which could be correlated with a particular teaching method or level of experience. Moreover, the sections taught by the author of the open book tests showed no significantly higher performance than the other sections.

The low average performance on these open book tests was revealing. It demonstrated that the average college freshman understood only about 60 percent of the material sampled from the text, and that the added instruction provided in laboratory and the presumably greater skill of experienced teachers did not improve this performance. (From experience on faculty committees, observing the frequency of misunderstandings, I am convinced that the efficiency of communication between faculty members also is less than 60 percent.)

One consistent correlation observed in this study was in the performance of individual students on successive tests. Those students who obtained high scores on the first test were usually able to repeat this performance on subsequent tests, and the students who scored low were usually unable to improve their performance on subsequent tests (see Verduin, 1950, for more detailed informa-

tion on open book objective tests). There was also a highly significant, positive correlation between the final grades given by each instructor and the performance of the students on open book tests, although several instructors gave only 10 percent weight to the open book test scores in determining student grades. The degree of correlation can be expressed by noting that if the instructors had based the grades entirely on the open book tests the students would, in 63 percent of the cases, have received the same letter grade as they actually did receive on the basis of all the evidence gathered by the instructor, and in less than 2 percent of the cases did the grade actually given differ by more than one letter from the grade indicated by the performance on open book tests. Tests of this type, therefore, can provide valuable criteria for grading the student.

The data in table 1 do not warrant the conclusion that biology students did not profit from laboratory experience, only that their ability to analyze open book objective type questions was not improved by it. Obviously an examination in which biological specimens were introduced, would place the laboratory group at a distinct advantage, and the evident intellectual satisfaction derived from laboratory experience is unquestioned.

One tangible result of this experiment was the rearrangement of the general biology curriculum to combine the nonlaboratory course with the laboratory course into a single one-semester Biology offering, providing one two-hour laboratory period and two one-hour discussion periods each week. This course now serves as a cultural course for those who plan no further study of biology and as a foundation course for those who plan to specialize in the life sciences. The evidence in table 1, that the students selecting the nonlaboratory course were not inferior to the others in their ability to understand biological concepts justified this amalgamation.

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- Verduin, J. 1950. An open book objective examination for science courses. *School Sci. and Math.* 50: 213-221.

Perspectives in Marine Biology. A symposium held at Scripps Institution of Oceanography, March 24-April 2, 1956. Papers by 41 authors, edited by A. A. Buzzati-Traverso, Univ. of California Press. 1958. 621 pp. \$10.00.

The author of the first paper in this book stated that, "We are asked to suggest what might be done to reach a better understanding of the relationships between organisms and between them and their physical environments, and how we may bring some of the newer and more exciting experimental techniques to bear on these problems."

In Dr. Rae's opinion, current studies will continue to pose problems rather than provide solutions because "... (1) until we can take a closer look at the animals and learn more of their intimate biology our field work will lack objectivity; (2) unless our laboratory work can become more realistic, it will not necessarily be relevant to the behavior of the animals in their normal habitats." He concluded his consideration of "Parameters of the Marine Environment" by reemphasizing the point that we are not well prepared to take advantage of the excellent techniques of the experimentalists.

The second paper, by Dr. Alfred C. Redfield, was entitled "The Inadequacy of Experiment in Marine Biology," and its theme is the greater need now for more knowledge of the phenomena to be explained rather than for experimentation.

The papers are grouped under subject headings in four parts, as follows: Ecology, Physiology and biochemistry, Behavior, and Genetics and evolution. The authors are well-known scientists, and most of the papers led to comments by members of the group. The final paper, by the editor, presents a digest of ideas expressed during the sessions of the "idea groups" as suggestions for further development of marine biology.

The series of papers is well assembled and is stimulating reading for all scientists whose interests pertain to marine biology.

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